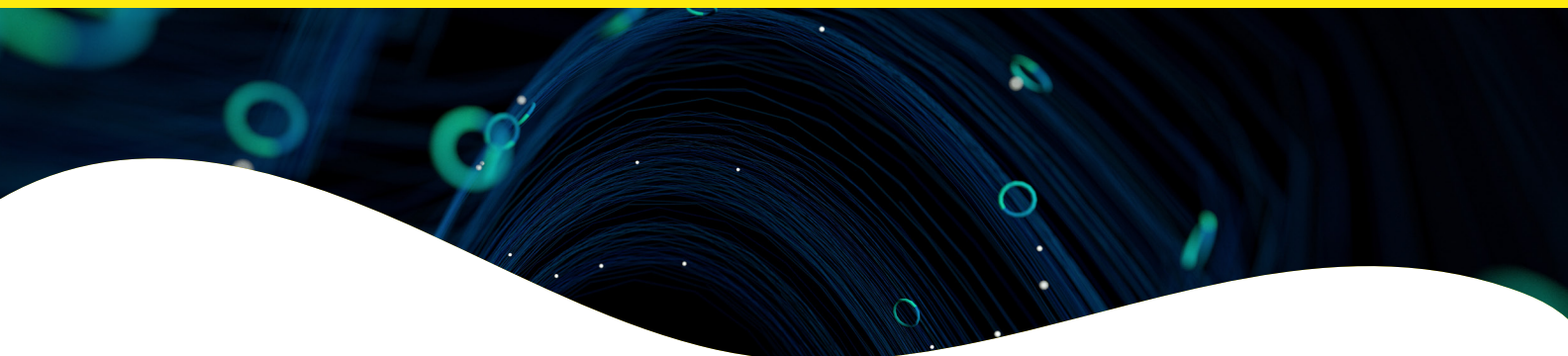


# HTRF setup recommendations for SpectraMax i3.



## HTRF Europium cryptate donor / red acceptor readout setup recommendations for SpectraMax i3

To read HTRF™, the SpectraMax i3® must be first equipped with the SpectraMax i3 Revvity HTRF cartridge, which enables the simultaneous measurement of both 620 nm donor and 665 nm acceptor emissions. 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.

HTRF readout can be achieved by SpectraMax i3 readers after the installation of the HTRF dedicated cartridge, which includes the optimized excitation and emission filters, the light source and the dichroic mirrors. The measurement conditions should then be set up in the SoftMax Pro software according to the following indications:

| Setup                        |  |
|------------------------------|--|
| Cartridge                    | HTRF Detection cartridge   |
| Number of flashes            | 30   |
| Integration delay (lag time) | 70 µs  |
| Integration time             | 400 µs   |
| Optimal z-position           | Volume and plate format dependant.<br>Must be optimized before each new configured measurement using the labware optimization procedure of the software. |

## HTRF Terbium cryptate donor / green acceptor readout setup recommendations for SpectraMax i3

To read HTRF, the SpectraMax i3 must be first equipped with the SpectraMax i3 Revvity HTRF cartridge, which enables the simultaneous measurement of both 620 nm donor and 520 nm acceptor emissions. 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.

HTRF readout can be achieved by SpectraMax i3 readers after the installation of the HTRF dedicated cartridge, which includes the optimized excitation and emission filters, the light source and the dichroic mirrors. The measurement conditions should then be set up in the SoftMax Pro software according to the following indications:

| Setup                        |  |
|------------------------------|--|
| Cartridge                    | HTRF Detection cartridge   |
| Number of flashes            | 30   |
| Integration delay (lag time) | 70 $\mu$ s   |
| Integration time             | 400 $\mu$ s  |
| Optimal z-position           | Volume and plate format dependant.<br>Must be optimized before each new configured measurement using the labware optimization procedure of the software Volume and plate format dependant. |

## HTRF Terbium cryptate donor / red acceptor readout setup recommendations for SpectraMax i3

To read HTRF, the SpectraMax i3 must be first equipped with the SpectraMax i3 Revvity HTRF cartridge, which enables the simultaneous measurement of both 620 nm donor and 665 nm acceptor emissions. 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.

HTRF readout can be achieved by SpectraMax i3 readers after the installation of the HTRF dedicated cartridge, which includes the optimized excitation and emission filters, the light source and the dichroic mirrors. The measurement conditions should then be set up in the SoftMax Pro software according to the following indications:

| Setup                        |  |
|------------------------------|--|
| Cartridge                    | HTRF Detection cartridge   |
| Number of flashes            | 30   |
| Integration delay (lag time) | 70 $\mu$ s   |
| Integration time             | 500 $\mu$ s  |
| Optimal z-position           | Volume and plate format dependant.<br>Must be optimized before each new configured measurement using the labware optimization procedure of the software Volume and plate format dependant. |

\*The fluorescence ratio is a correction method developed by Revvity with an application limited to the use of HTRF reagents and technology, and for which Revvity has granted a licence to Molecular Devices. The method is covered by the US patent 5,527,684 and its foreign equivalents.

