HTRF® Terbium cryptate donor / Green acceptor readout
Setup recommendations for Safire2TM

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.

Safire2TM readers must be appropriately configured for HTRF® readout by setting up the measurement conditions in the "multilabeling" function of Xfluor4 or Magellan 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:

Measurement 1

Excitation wavelength	343 nm
Excitation bandwidth	20 nm
Emission wavelength	620 nm
Emission bandwidth	10 nm
Number of reads	100
Lag time	60 µs
Integration time	500 µs
Gain	Optimal
Z position	Optimal

Measurement 2

Excitation wavelength	343 nm
Excitation bandwidth	20 nm
Emission wavelength	520 nm
Emission bandwidth	10 nm
Number of reads	100
Lag time	60 µs
Integration time	500 µs
Gain	Optimal
Z position	Optimal

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

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