Handbook
Hypoxia Probe Solution (LOX-1)

Please refer to expiration date on the label.

Precautions for use
Please read all the instructions before use.

Introduction
This probe is a phosphorescent light-emitting iridium complex. Phosphorescence of this probe is quenched by oxygen, and it is increased in response to low levels of oxygen which is detectable by a general fluorescent microscopy (red-fluorescence). The signal change is reversible, and this probe easily permeates cell membrane. So, the reagent shows real-time response according to oxygen levels in living cells. This probe can be used to observe hypoxia condition inside spheroid and detect subcutaneous xenograft tumors in small animals.

Spectral Characteristics
Absorption spectrum is given in green and phosphorescence in red. Absorption and phosphorescence are peaked at 483 and 616 nm, respectively.

Example of use for Hypoxia Probe Solution
1. Hypoxia Probe Solution (LOX-1) is stored at -20°C except for use.
2. Hypoxia Probe Solution is diluted with culture medium to prepare 20 μmol/L working solution just before use (100 times dilution).
3. Add 10 μL-aliquots of the working solution into 100 μL culture medium gently (final concentration of the probe is 2 μmol/L).
4. After incubation for one day, red-phosphorescence is observed with a general fluorescent microscopy with the standard TRITC filter set (for example G-2A filter block: Ex 510-560, DM575, BA590).

**Note:** Incubation time depends on your sample. For monolayer cells, incubation time may be enough 0.5 hrs. For spheroids, it may be 8-12 hrs.

**Note:** We recommend using FITC excitation and Texas Red® emission filters for best results.

**Reference**

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