



Revised: December 12, 2011

# **Product Information**

# RedDot™2

Catalog #	Unit Size	Concentration	Storage
40061-T	25 µl		Store at 4°C, protect from light
40061	250 µl	200X in DMSO	
40061-1	1 mL		nonniight

#### Storage and Handling

Store RedDot2 at 4°C, protected from light. RedDot2 is chemically stable and can tolerate multiple freeze/thaw cycles. When stored as directed, RedDot2 is stable for at least one year from the date of receipt.

CAUTION: RedDot2 may be toxic and mutagenic. Handle with care. Dispose of RedDot2 as toxic waste according to your institution's regulations.

#### Spectral properties

Abs/Em maxima with DNA: 665/695 nm

RedDot2 can be excited at a wide range of wavelengths including the 488 nm laser for flow cytometry and by wavelengths up to 647 nm.

## **Product Description**

RedDot2 is a cell membrane-impermeable, far-red dye with high selectivity for membrane compromised or dead cells. RedDot2 staining is nuclear specific in fixed and permeabilized cells and tissue sections and does not require RNase treatment.

RedDot2 is highly thermostable and photostable. It does not require a wash step and demonstrates greater photostability than the traditional blue fluorescent counterstain DAPI. RedDot2 can be excited by wavelengths from 488 to 647 nm and emits far red fluorescence with emission maximum at 695 nm.

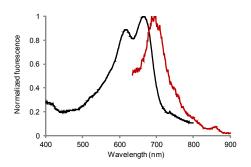


Figure 1. Absorption and emission spectra of RedDot2 with DNA.

#### **Staining protocols**

#### For selective staining of dead cells

- 1. Dilute RedDot2 to a final concentration of 1X in cell culture medium.
- Add cell culture medium containing RedDot2 to cells and incubate 10 minutes at room temperature.
- Detect far red nuclear staining by fluorescence microscopy, flow cytometry, or fluorescence plate reader.

# For nuclear staining of fixed and permeabilized cells or tissue sections

- Fix cells according to your standard protocol (e.g. 4% paraformaldehyde in phosphate buffered saline (PBS) for 15 minutes at room temperature, or chilled methanol for 5 minutes at -20°C).
- 2. Rinse cells twice with PBS.
- Permeabilize cells in 0.5% Triton X-100 in PBS for 10 minutes at room temperature.
- 4. Rinse cells twice with PBS.
- Optional: perform immunofluorescence staining according to your standard protocol.
- Dilute RedDot2 in PBS to a final concentration of 1X. Stain cells for 10-30 minutes at room temperature. Note: RedDot2 also can be diluted in immunofluorescence blocking buffer or incubated together with antibodies during immunofluorescence staining.
- Optional: rinse cells with PBS and mount in fluorescence antifade mounting medium.
- 8. Detect far red nuclear staining by fluorescence microscopy, flow cytometry, or fluorescence plate reader.

### **Related Products**

Cat.#	Product Name	Unit Size
40060-T	RedDot1	25 µl
40060	RedDot1	250 µl
40061-1	RedDot1	1 ml
23001	EverBrite Mounting Medium	10 ml
23002	EverBrite Mounting Medium with DAPI	10 ml
40009	DAPI, dilactate	10 mg
40011	DAPI (dihydrochloride salt)	10 mg
40043	DAPI in water, 10 mg/ml	1 ml
40016	Propidium iodide	100 mg
40017	Propidium iodide in water, 1.0 mg/ml	10 ml
40048	Propidium iodide buffer, 50 µg/ml	2 ml
40044	Hoechst 33258 in water, 10 mg/ml	10 ml
40045	Hoechst 33258, pentahydrate	100 mg
40046	Hoechst 33342 in water, 10 mg/ml	10 ml
40047	Hoechst 33342, trihydrochloride trihydrate	100 mg

Materials from Biotium are sold for research use only, and are not intended for food, drug, household, or cosmetic use.