

Nuclear stains

For fluorescence microscopy

RedDot™, NucSpot® and other nuclear dyes

NucSpot® Live 470

RedDot[™] dyes are far-red nuclear counterstains. Their fluorescence emission is well separated from the emission peaks of red, green and blue fluorescent probes, making RedDot[™] dyes ideal for multicolor imaging.

RedDot™1

Cell permeable RedDot[™]1 stains the nuclei of live cells rapidly and specifically (Figure 1). It also has been used to stain nuclei in live organisms¹. It can also be used for cell cycle analysis in live cells by flow cytometry (Figure 3). Note: Similar to Draq5[™], RedDot[™]1 shows cytotoxicity within 4 hours of staining. For long term live cell imaging experiments, we recommend our NucSpot® Live Stains (see other side).

RedDot™2

Cell membrane-impermeable RedDot[™]2 has excellent selectivity for dead cells. Our NucView®488 and RedDot[™]2 Apoptosis & Necrosis Kit pairs RedDot[™]2 with NucView®488 caspase-3 substrate for detection of apoptotic and necrotic cells. Unlike Draq5[™] and Draq7[™], which show significant cytoplasmic staining in permeabilized cells unless additional blocking steps are performed, RedDot[™]2 staining is nuclear-specific. Thus, RedDot[™]2 provides excellent nuclear counterstaining in fixed and permeabilized cells (Figure 2).

RedDot™1

- · Far-red fluorescence
- Cell-permeable, for staining live cells
- Can be used for cell cycle analysis by flow cytometry

RedDot™2

- · Far-red fluorescence
- · For fixed cells or tissues, or selective dead cell staining
- Nuclear-specific, no RNAse or wash required

Ordering information

Cat. #	Product name	Size
40060-1	RedDot™1 Far-Red Nuclear Stain, 200X in H ₂ O	1 mL
40060		250 uL
40060-T		25 uL
40061-1	RedDot™2 Far-Red Nuclear Stain, 200X in DMSO	1 mL
40061		250 uL
40061-T		25 uL
30072	NucView® 488 and RedDot™2 Apoptosis and Necrosis Kit	100 assays

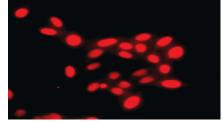
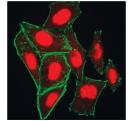


Figure 1. Live HeLa cells stained with RedDot™1 for 5 minutes at 37°C.

RedDot™2 vs. Draq7™



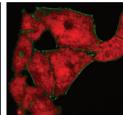


Figure 2. Fixed and permeabilized HeLa cells stained with RedDot™2 (left) or Draq7™ (right). RedDot™2 staining is highly selective for the nucleus, while Draq7™ stains the nucleus and cytoplasm unless a separate blocking step is performed.

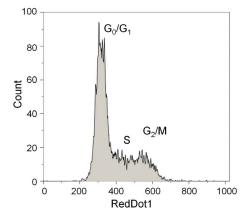


Figure 3. RedDot[™]1 staining for cell cycle distribution analysis. Live Jurkat cells were stained with RedDot[™]1, then analyzed by flow cytometry with 633 nm excitation and 710/50 BP emission filter. Image courtesy of Philip Hexley, Shriners Flow Cytometry Core Facility, Shriners Hospital for Children and University of Cincinnati.



NucSpot® 470 Nuclear Stain for dead or fixed cells

NucSpot® 470 is a cell membrane-impermeant green fluorescent DNA stain. It is virtually non-fluorescent in the absence of DNA, but fluoresces bright green upon DNA binding. While other green nucleic acid stains like TOTO®, TO-PRO®, or SYTOX® dyes stain both the nucleus and cytoplasm, NucSpot® 470 specifically stains the nucleus of fixed and permeabilized cells or tissue sections (Figure 4). It also can be used to selectively stain dead cells in living cultures. It has green fluorescence that can be imaged using standard settings for FITC. With excitation at 460 nm, it also is an excellent match for instruments with blue LED excitation sources.

NucSpot® Live Cell Nuclear Stains for live or fixed cells

NucSpot® Live Cell Nuclear Stains are cell-membrane permeable DNA dyes that specifically stain nuclei in live or fixed cells. They have excellent specificity for DNA without the need for a wash step, and they have low toxicity for live cell imaging. The dyes are supplied as 1000X stock solutions in DMSO, and are supplied with a vial of verapamil, which may increase probe retention and staining in some cell types.

NucSpot® Live 488 stains the nuclei of live or fixed cells with green fluorescence. NucSpot® Live 650 stains the nuclei of live or fixed cells with far-red fluorescence (Ex/Em 650/675 nm) for detection in the Cy®5 channel (Figure 5). The NucSpot® Live 650 fluorophore is also compatible with super-resolution imaging by SIM or STED. Note: NucSpot® Live 488 and NucSpot® Live 650 have dim blue fluorescence in the DAPI channel, and may not be suitable for multicolor imaging with blue probes.

NucSpot® Advantages

- Excellent nuclear specificity, no RNase or washing required
- · Green or far-red fluorescence
- NucSpot® 470 stains dead or fixed cells
- NucSpot® Live dyes have low toxicity, ideal for live-cell imaging
- NucSpot® 650 fluorophore is compatible with SIM or STED

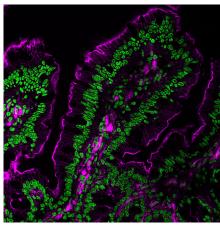


Figure 4. Intestine section stained with CF®640R phalloidin (magenta) and NucSpot® 470 (green).

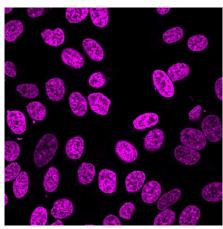


Figure 5. Live HeLa cells stained with NucSpot® Live 650 and imaged in the Cy® 5 channel.

Ordering information

Cat. #	Product name	Size
40081	NucSpot® 470 Nuclear Stain, 1000X in DMSO	50 uL
40081-T		10 uL
40081	NucSpot® 488 Live Cell Nuclear Stain, 1000X in DMSO	50 uL
40081-T		10 uL
40082	NucSpot® 650 Live Cell Nuclear Stain, 1000X in DMSO	50 uL
40082-T		10 uL
40046	Hoechst 33342, 10 mg/mL in H2O	10 mL
40044	Hoechst 33258, 10 mg/mL in H2O	10 mL
40047	Hoechst 33342, trihydrochloride trihydrate	100 mg
40045	Hoechst 33258, pentahydrate	100 mg
40043	DAPI in H2O, 10 mg/mL	1 mL
40011	DAPI	10 mg
40009	DAPI, dilactate	10 mg
23002	EverBrite™ Mounting Medium with DAPI	10 mL
23004	EverBrite™ Hardset Mounting Medium with DAPI	10 mL
40016	Propidium iodide (PI)	100 mg
40017	Propidium iodide, 1 mg/mL in H ₂ O	10 mL
40048	Propidium iodide buffer, 50 ug/mL in PBS	2 mL

Classic blue nuclear stains

Biotium also sells classic nuclear dyes at a competitive price

Hoechst:

Membrane-permeant blue dye for live or fixed cells

DAPI:

- Blue counterstain for fixed and permeabilized cells
- Available in EverBrite[™] antifade mounting media

Propidium Iodide (PI)

- Selective dead cell stain
- Used for cell cycle analysis by flow cytometry

