DRAQ5® Fluorescent Probe

1．规格： 5mM 50 μL（sufficient for ~4 × 96 wells），200 μL（200μL, sufficient for ~16 × 96 wells）

2．参数： MW~413 Da； Wavelength: Ex/Em= 646/697nm

3．储存： 4℃避光保存 2 年； 常温运输

4．产品描述：

DRAQ5® 是一种对双链 DNA 具有高亲和力的远红外荧光活细胞 DNA 染料。它是一种可以透过细胞膜的染料，可标记活细胞或固定后/死细胞。在流式细胞术中，这种染料可用于区分有核和无核细胞。由于 DRAQ5®能够按照化学计量比结合至 DNA 因此还可用于报告细胞核 DNA 含量，适用于染色体倍数和细胞周期分析。在荧光显微镜分析中，它可用作细胞核复染剂。DRAQ5 有很多应用，高度兼容现有仪器平台广泛使用的程序，主要的应用领域 HCS，细胞模型，GFP，流式细胞仪和荧光显微镜。

DRAQ5 激发波长范围为 488～647nm。对于成像显微术，建议使用 633 或 647nm 的光源进行激发。对于流式细胞术，在 488nm 处激发这种染料时，可使用 685LP 二向分色镜和 710/50 通道进行检测；在 633nm 处激发时， 可以使用 660/20 通道进行检测。对于细胞周期/DNA 分析应用，建议使用波长较长的滤光片，例如 735 LP 二向分色镜和 780/60 通道来优化 G1 和 G2/M 峰的 CV 值。请确保您的仪器能够检测该染料。

由于 DRAQ 发射和激发波长范围很宽，不建议将 DRAQ5 与其他可被 488 或 633 nm 激发的远红光荧光染料联用。

5．操作说明

（1）说明：在实验时，DRAQ5®是作为最后一种染料来染色的， 因为 DRAQ5®染色完不需要其余的清洗步骤，因此 DRAQ5 可以直接加在含有细胞的培养基中进行活细胞染色

（2） 操作

①. 准备不含有叠氮化钠的 PBS 缓冲液或特定细胞的特定 培养基。

②. 用 PBS 或培养基重悬细胞，控制细胞密度≤ 4 × 105 cells/mL。对于贴壁细胞和部分组织，大致估计细胞个数。

③. 按下表 1 加入相应体积的合适浓度的 DRAQ5 染色液， DRAQ5 染色液可以直接加到组织或者贴壁细胞的表面，或者直接加入到新鲜培养基中

④. 轻轻混匀，室温下孵育 5–30 min。37℃ 孵育，时间缩短 为 1–3 min。对于时间跨度较长的实验，`例如 EGFP 实验，DRAQ5 染色液要在激动剂和颉颃剂加入前的实验进程中（通常 0.5～3 h）加到培养基中，浓度控制在 1μM。注：如果在 DRAQ5 染色前，细胞已经被别的荧光染料染色，注意上述操作过程要避光。

⑤. 染色细胞可直接进行相应分析，不需要清洗等别的操作。 下表 1 细胞数目及所需 DRAQ 5 体积及终浓度。

DRAQ5® Fluorescent Probe

DRAQ5® Fluorescent Probe, 5mM, 50μL, sufficient for ~4 × 96 wells

DRAQ5® Fluorescent Probe, 5mM, 200μL, sufficient for ~16 × 96 wells

MW~413 Da, Wavelength: Ex/Em= 646/697nm

Features

- Stable at room temperature
- Rapid staining of cells without wash steps
- No UV excitation and no emission spectral overlap with PE

Storage&Shipping:

Store at 4°C protected from light. Product is shipped at ambient temperature.

Discription:

The DRAQ5® Fluorescent Probe is a far-red DNA stain for use in live or fixed cells. Because of its far-red excitation and emission, the DRAQ5 Fluorescent Probe can be multiplexed with many other fluorophores and is ideal for cells expressing green fluorescent protein (GFP) fusion proteins. DRAQ5 Fluorescent Probe is compatible with many existing protocols across a wide range of instrumentation platforms. This fluorescent probe can be used for cellular imaging.

Fluorescence cell-based assays, such as flow cytometry, in-cell ELISA, fluorescence microscopy and high-content imaging require a fluorescent label to identify individual cells. When using multiple fluorescent probes to detect different cellular targets or activities, each probe must have a fluorescent spectrum different than the other probes. The blue-fluorescent DNA- binding probes, Hoechst and DAPI are frequently used; however, these probes cannot be used when UV illumination is unavailable or other blue-emitting fluorescent probes are used. Therefore, nuclear probes that emit in a color other than blue are useful for cell identification and counting, and for determining nuclear morphology and DNA content.

The DRAQ5® Fluorescent Probe emits in the far-red region, is lipophilic and crosses cell and nuclear membranes in live and fixed cells and tissues for rapid DNA staining. This stain is water-soluble, supplied ready to use and does not require RNase, cell lysis, or a washing step, making it compatible with automation. Because DNA staining is stoichiometric, the DRAQ5 Fluorescent Probe can be used for DNA content analysis in cell proliferation studies.

Procedure for DNA Staining

Note: DRAQ5® Fluorescent Probe is usually added as the last stain in a labeling procedure because no washing is required. Alternatively, add this probe in assay medium for a live cell assay. Read the supplied material safety data sheet before handling DRAQ5® Probe.

1. Prepare phosphate-buffered saline (PBS, without sodium azide) or the appropriate culture media for the specific cells.
2. Resuspend cells in PBS or media at ≤ 4 × 105 cells/mL in a test tube. For adherent cells estimate the number of cells based on confluence level or tissue section dimensions.
3. Add DRAQ5 Fluorescent Probe directly as supplied according to the volumes indicated in Table 1. Add directly on top of tissue sections and adherent cells or add DRAQ5 Fluorescent Probe in fresh media.
4. Gently mix and incubate for 5–30 minutes at room temperature. DRAQ5 Fluorescent Probe staining is accelerated at 37°C and may be reduced to 1–3 minutes. For time-lapsed assays (e.g., studying translocation of an EGFP-tagged protein) DRAQ5 Fluorescent Probe may be added to the assay media for the duration of the assay (typically 0.5–3 hours) at 1µM before adding any agonist/antagonist.

Note: Protect cells from light during incubation if other (immuno-) fluorescent stains have been applied to the cells before the DRAQ5 Fluorescent Probe staining.

5. Cells can be analyzed directly without further treatment or washing.

Cell sample preparation:		VOLUME OF DRAQ5® (AS SUPPLIED) REQUIRED FOR A CONCENTRATION OF:		
No. of cells:	in volume:	5 µM	10 µM	20 µM
1 x 106	2500 µl	2.5 µl	5 µl	10 µl
4 x 105	1000 µl	1 µl	2 µl	4 µl
2 x 105	500 µl	0.5 µl	1 µl	2 µl
1 x 105	250 µl	0.25 µl	0.5 µl	1 µl
5 x 104	125 µl	0.13 µl	0.25 µl	0.5 µl