

## Data Sheet

# BioTracker™ BioCyTASQ G-quadruplex (G4)

## Cell Probe

**SCT246**

**Pack Size: 1 mg**

**Store at -20 °C**

**FOR RESEARCH USE ONLY**

**Not for use in diagnostic procedures. Not for human or animal consumption.**

## Background

G-quadruplexes are four-stranded structures comprising stacked G tetrads (G4) formed within certain guanine-rich nucleic acid sequences. G-quadruplexes are enriched in open chromatin regions but are also present in the nucleolus and cytoplasm. The formation of G4 structures is dynamic and cell-type specific, suggestive of interaction with transcription factors and other proteins.<sup>1</sup> For this reason G-quadruplexes have garnered intense interest for their potential functions in transcriptional regulation.

The BioTracker™ BioCyTASQ G-quadruplex G4 Cell Probe is optimized for cellular detection of G-quadruplexes. The BioCyTASQ probe utilizes the template-assembled synthetic G-quartets (TASQ) concept as a biomimetic ligand for G-quadruplexes, a design which results in high specificity for folded G4. BioCyTASQ is water-soluble and demonstrates robust intracellular accessibility. BioCyTASQ detects ribosomal RNA-G4 associated with perinuclear rough ER regions to a greater degree compared to BG4 antibody. The biotin moiety on the BioCyTASQ molecule enables detection with streptavidin. BioCyTASQ is suitable for G4 capture pull-down assays as well as cellular imaging.<sup>2</sup> BioCyTASQ G4 Cell Probe is not recommended for live cell imaging applications. The optimized design of the BioTracker™ BioCyTASQ G-quadruplex G4 cell probe makes it a powerful tool for biochemical and cellular analysis of G4.

## Source

BioTracker™ BioCyTASQ G-quadruplex G4 Cell Probe (SCT246) does not contain genetically modified organisms.

## Spectral Properties

This probe is non-fluorescent. Probe may be imaged using fluorophore-conjugated streptavidin.

## Quality Control Testing

Purity: ≥ 98% confirmed by HPLC, HNMR, LC-MS and elemental analysis

Molar Mass: 968 g/mol

## Storage and Handling

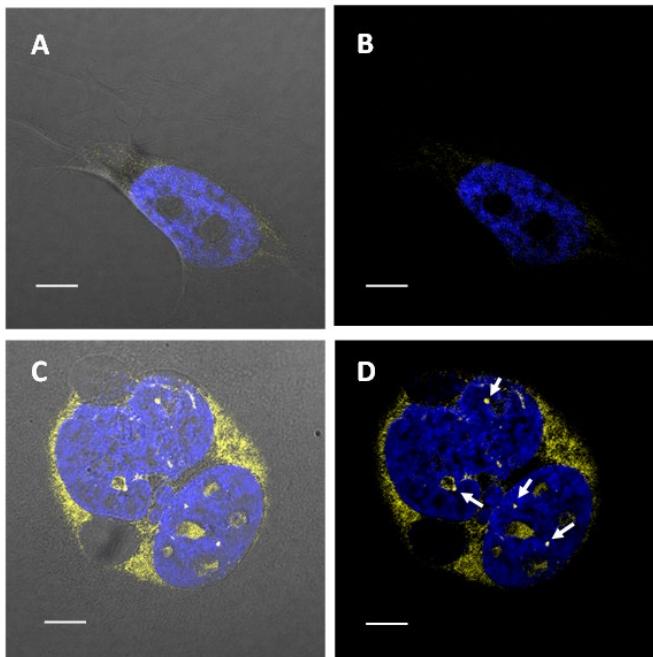
Store BioTracker™ BioCyTASQ G-quadruplex G4 cell probe at -20 °C, desiccated and protected from light.

**Note:** Centrifuge vial briefly to collect contents at bottom of vial before opening.

## Presentation

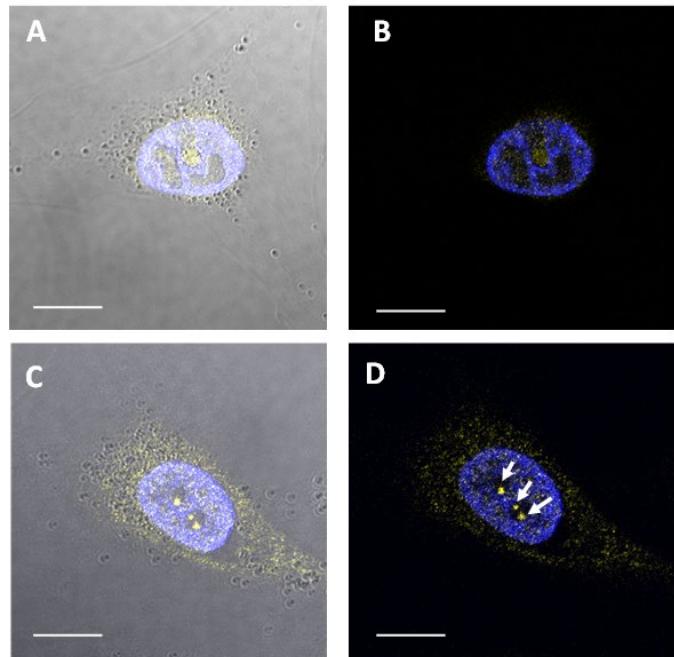
Lyophilized. White solid

## Representative Data



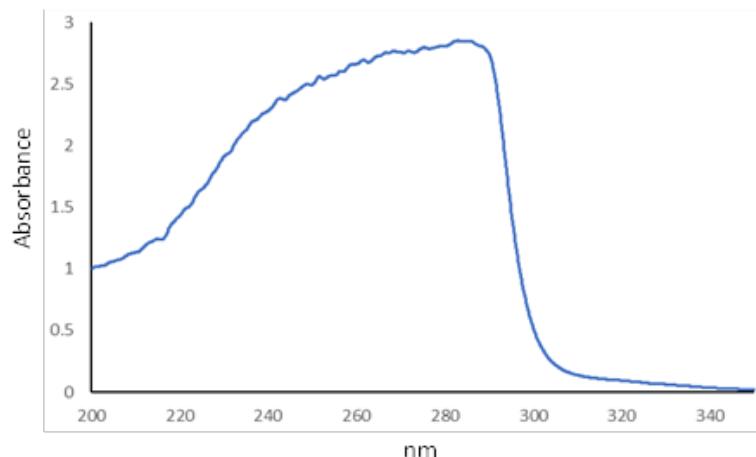
**Figure 1.** Confocal microscopy images of G-quadruplexes detected with BioCyTASQ. Paraformaldehyde-fixed HeLa cells were incubated without (**A, B**) or with BioCyTASQ (**C, D**), then incubated with Cy3-conjugated streptavidin (S6402, yellow), co-stained with DAPI nuclear dye (90225, blue) and merged together. (**A, C**) overlay with brightfield images. White arrows indicate location of G4 foci. Perinuclear staining is associated with RNA-G4 in rough endoplasmic reticulum.

Scale bars: 10  $\mu$ M



**Figure 2.** Confocal microscopy images of G4 foci in HeLa cells detected with anti-DNA G-quadruplex structure BG4 antibody (MABE917) and anti-FLAG antibody (MAB3118, yellow), co-stained with DAPI nuclear dye (blue). No BG4 control (**A, B**) versus BG4 stained cells (**C, D**). (**A, C**) overlay with brightfield images. White arrows indicate G4 foci in nucleus.

Scale bars: 10  $\mu$ M



**Figure 3.** Absorption spectrum of SCT246 BioCyTASQ in water.

## Protocols

### Preparing BioTracker BioCyTASQ G4 cell probe stock solution

1. Before opening the vial, spin down the solid to the bottom by a microcentrifuge or by a desktop centrifuge.
2. Warm the vial to the room temperature. Prepare the BioCyTASQ (Molecular Weight: 968 g/mol) probe stock solution by dissolving the contents of one vial (1 mg) in 968  $\mu$ L of water to create a 1 mM solution.
3. Aliquot and store stock solution at  $-20^{\circ}\text{C}$  or below for longer-term storage.

### Labeling cells

1. Culture cells in an appropriate medium and vessel for fluorescence microscopy.
2. Prepare the BioCyTASQ staining solution by diluting the BioCyTASQ stock solution 1:1000 in PBS (1  $\mu\text{M}$  final concentration).
3. Fix cells in 4% paraformaldehyde.
4. Permeabilize cells with PBS + 0.1% (v/v) Triton<sup>®</sup> X-100 for 10 minutes at ambient temperature.
5. Add sufficient BioCyTASQ staining solution to cover the cells and incubate for 1 hour at ambient temperature.
6. Wash cells 3 times for 5 minutes with PBS.
7. Fix cells (for example, ice-cold methanol for 10 minutes).
8. Wash cells 3 times for 5 minutes with PBS.
9. Incubate cells with 1  $\mu\text{g}/\text{mL}$  fluorophore-conjugated streptavidin (for example, Cy3-streptavidin, S6402) in PBS for 1 hour at ambient temperature.
10. Wash cells for 5 minutes with PBS. If desired, counterstain with DAPI (1  $\mu\text{g}/\text{mL}$ ) for 10 minutes at ambient temperature.
11. Image cells on fluorescence microscope at wavelengths appropriate for fluorophores used.

**Note:** Optimal concentration must be determined by end user.

## References

1. Nat. Chem. 2021, 13(7): 626-633.
2. ACS Chem Biol. 2021, 16(5): 905-914.

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