

# RASTRUM

## *Protocol*

Incorporation of DQ™ Collagen Type I Bovine Skin, Fluorescein Conjugate into RASTRUM™ Matrix



## Introduction

DQ™ Collagen Type I Fluorescein Conjugate can be used to directly monitor collagenase activity of the cells encapsulated in RASTRUM advanced cell models. The generation of fluorescein signals is driven by the enzyme-driven hydrolysis of the substrate that separates the dye molecules from one another.

## Equipment and reagents required, but not provided

- DQ™ Collagen Type I From Bovine Skin, Fluorescein Conjugate (ThermoFisher: D12060)
- Sodium azide
- Deionised (DI) water
- 50°C waterbath
- Pipette (20, 200, 1000 µL) and pipette tips
- Sterile syringe
- 0.22 µm syringe filter

## Protocol

1. Prepare at least 2 mL, sterile 2 mM sodium azide solution in DI water.
2. Add 1 mL of the sterile 2 mM sodium azide solution into the 1 mg powder of DQ™ Collagen Type I From Bovine Skin, Fluorescein Conjugate to make a stock solution at 1 mg/mL.
3. Mix the solution using pipette until homogenous. If necessary, apply gentle heating (up to 50°C) and/or agitate the sample in a water bath for roughly 5 min.
4. Cover this DQ Collagen stock solution in aluminum foil and store at 4°C away from light.

*Note: Do not freeze the DQ Collagen stock solution.*

5. Establish the concentration of DQ Collagen substrates desired in the activator.

*Note: You can have up to 25 µg/mL DQ Collagen in the cell model.*

6. Calculate the volume of the DQ Collagen stock solution to be transferred to the activator according to the following formula:

$$\text{Stock Solution Volume } (\mu\text{L}) = \text{Target concentration } (\mu\text{g/mL}) \times \text{Activator Volume (mL)} \times 2$$

*Note: For example, if you would like to have a DQ Collagen concentration of 25 µg/mL in the cell model, you need to transfer  $25 \times 0.2 \times 2 = 10 \mu\text{L}$  of the stock solution to 0.2 mL activator.*

7. Pipette the calculated Stock Solution Volume of the DQ Collagen stock solution into the thawed activator. Mix well by pipetting the solution up and down for at least 5 times.
8. The resulting activator is ready to be mixed with your cells and to continue the PrintRun process according to your PrintRun protocol.



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