

Jun 25, 2024

Lysosomal activity DQ-Red BSA assay

DOI

<https://dx.doi.org/10.17504/protocols.io.j8nlk8dp6l5r/v1>

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Protocol Citation: Md. Razaul Karim 2024. Lysosomal activity DQ-Red BSA assay. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.j8nlk8dp6l5r/v1>

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Protocol status: Working

We use this protocol and it's working

Created: June 07, 2024

Last Modified: June 25, 2024

Protocol Integer ID: 102397

Keywords: Lysosomal activity DQ-Red BSA assay, Cell culture, Imaging, protocol details the lysosomal activity dq, lysosomal activity dq, red bsa, assay

Funders Acknowledgements:

ASAP

Grant ID: 000592

Abstract

This protocol details the Lysosomal activity DQ-Red BSA assay.

Materials

▪ Reagent:

DQ-BSA Red (Thermo Fisher Scientific)

▪ Coverslips:

Poly-D-Lysin coated coverslips. Corning BioCoat (Ref: 354086).

 Corning® BioCoat™ Poly-D-Lysine #1 German Glass Coverslip **Merck Catalog #354086**



Cell Culture/Treatments:

1h 30m

1 0.5×10^4 /ml of cells plated on coverslips.

2 PFF-488 treatment.

3 PBS/trypsin (0.01%) wash.



4 **DQ-BSA Red treatment:**

1h 30m

10 μ L 01:30:00 at 37 °C .



5 Fix cells using 4% PFA.

Imaging:

6 Red fluorescence signal is detected only when fluorogenic substrate is hydrolyzed by lysosomal proteases.

7 Leica Stellaris 8 confocal laser scanning microscope.

8 **Image analysis:**



Image J. quantification of the % area of the cell covered by DQ-Red signal divided by the number of cells (DAPI stained).