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Lysosomal activity DQ-Red BSA assay

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Razaul Karim¹

¹University of Minnesota, Team Lee

ASAP Collaborative Rese...

Team Lee



Jane Balster

ASAP - Team Lee





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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 0.5x10⁴/ml of cells plated on coverslips.
- 2 PFF-488 treatment.
- 3 PBS/trypsin (0.01%) wash.



4 DQ-BSA Red treatment:





5 Fix cells using 4% PFA.

Imaging:

- Red fluorescence signal is detected only when fluorogenic substrate is hydrolyzed by lysosomal proteases.
- 7 Leica Stellaries 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).