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DQ-BSA assay

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Protocol status: Working

We use this protocol and it's working

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Abstract

This protocol describes the DQ BSA assay for measuring the lysosomal proteolytic activity.

Attachments



[mqjxbj3up.docx](#)

15KB

Materials

Reagents

1. DQ BSA - #D12051 (1mg/ml prepared in sterile PBS), working concentration 10µg/ml.
2. BSA-488 - #A13100 (5mg/ml prepared in sterile PBS), working concentration 5 µg/ml.
3. Mattek glass bottom dishes



DQ-BSA assay

3h 10m

- 1 Plate 100,000 mature macrophages or microglia or BMDM were seeded on Mattek glass bottom dishes.
- 2 Next day treat cells with IMJ 50 nanomolar (nM) MLI2 or IMJ 250 nanomolar (nM) LRRK2-IN-1 for 🕒 02:00:00 followed by 🕒 01:00:00 treatment with 🧴 10 µg/mL DQ-BSA (Thermo Fisher Scientific, #D12051) and 🧴 50 µg/mL Alexa-488 BSA (Thermo Fisher Scientific #A13100) in the media containing inhibitors.
- 3 After 3 hours wash the cells 3X with culture media. 🧴
- 4 Then add 🧴 1 mL culture media to the washed cells and return to incubator for 🕒 00:10:00 . 🧴 📷 10m
- 5 Imaging was done on LSM 880.