Scintillation Count of radiolabeled whole cell

Elizabeth Fozo[1]
[1]In-house protocol

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1 Works for me This protocol is published without a DOI.

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ABSTRACT

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ABSTRACT

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Steps

1 Grow OG1RF to mid log phase in BHI.

2 Harvest 5mL of cells.

3 Wash twice in 1X davis salts without glucose.

4 Re-suspend in 5mL 1X davis salt without glucose with 10mg/mL BSA.

5 Incubate at 37^{O}C for 30 minutes as starvation treatment.

6 Move to hot room.

7 Place cells in 37^{O}C heat block for assay or in 95^{O}C heat block for certain death.
- Prior to assay I make a stock of hot/cold OA.

8 Add the hot/cold mix to 5mL of cells in the davis/BSA buffer and start the timer.

9 Immediately take the 200ul of cells out for T0 - there are placed on the filter paper which is connected to a vacuum - these are washed with 2 mL of wash buffer (1X davis salts with 10mg/ml BSA).

10 Stager the time points every 5 minutes for about 20-25 minutes. Then , repeat this for the heat killed cells. Note that there isn't any difference between the heat killed cells and the 37^{O}C cells. We are still working on control for this part of the assay.

11 At completion of the assay filters are added to vials with scintillation fluid and counted by the machine.