

Exp. wB5001

9/24/01

$^3\text{HTdR}$ Preparation: 10 ml D medium + 200 μL $^3\text{HTdR}$ stock
yields 20 kCi/ml D medium

8:20 am seed flasks w/ cells, 37°C 2.5% CO_2

11:20 am added 1 ml tritium 20 kCi/ml - final conc. 4 kCi/ml
returned to incubator 37°C 2.5% CO_2

2:20 pm remove flask 1, wash 7X w/ 5 ml PBS/PS

trypsinize w/ 0.5 ml for 4 min

add 1.5 ml PBS/PS to resuspend & transfer to 14 ml tube

syringe 5X with 3 ml 2LG w

transfer 3 x 200 μL to 7 ml scintillation vials

and add 5 ml Ecolume

transfer 1 x 100 μL to Coulter cup & count with
200 μL IsotoneTM
500 μL manifold setting

Coulter 1800 1788 1776

centrifuge remaining cells at room temp, 1500 rpm, 5 min

transfer 3 x 200 μL supernatant to 7 ml scintillation vials

and add 5 ml Ecolume

4:20 pm remove flask 2, repeat above procedure

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9:50 pm remove flask 3, repeat above procedure (~50% confluent)

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9/25 12:20 am remove flask 4, repeat above procedure (~100% confluent?)

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