DNT Detection In Soil

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1 The 1% bacteria (PYB1s-yqjF3rd-luxCDABE) were cultured in LB containing 0.1% streptomycin (50 mg/mL) for 24 h with aeration at 37°C to reach a final density of 1 × 108 cells/mL.

2 Preparation of 2% Alginate solution, 1% calcium-chloride solution, experimental soil:dissolve 2 g Alginate in 100 mL distilled water (2% w/w) and 1g calciumchloride in 100 mL distilled water (1% w/w). The experiment soil was filtrated prior to use.

3 Sterilize the 2% Alginate solution, 1% calcium-chloride solution, (121°C, 15min), and experimental soil (121°C, 1.5h).

4 Add 0, 0.25, 0.5 and 1 mg of DNT powder (0, 250, 500 and 1000 mg/kg) to the wells of a 12-well plate.Take 1g of experimental soil and drop it onto the DNT in the 12-well plate on a clean bench.

5 The engineered bacteria were added into 2% Alginate solution at a 1:9 (v/v) ratio.

6 Drop 200 µL of 1% (w/w) calcium-chloride solution into the 1 mL alginate-bacterial suspension droplets (the volume ratio between the Alginate mixture and calcium chloride solution was 5:1). Leave the mix for 1 minute.

7 The mixed Alginate gel was tiled on the soil.

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