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Pour plating protocol for Emiliania huxleyi (single colonies)

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Protocol status: Working
We use this protocol and it's working

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Abstract

A protocol for generating single colonies of Emiliania huxleyi embedded in soft-agarose.

1 Autoclave 75 mL DDW with 0.75 g agarose

2 For 300 mL (= 7 plates): heat 225 mL FSW in a 50 °C water bath.

3 Cool autoclaved agar to 50 °C, mix with heated FSW, add F/2 stock and antibiotics if necessary. Swirl/mix well.

4 For each sample, pour 40 mL of FSW-agar mixture to a 50 mL conical tube.

5 Move to 32 °C bath.

6 Prepare the cells at desired cell concentration. For each sample prepare 0.5ml cells.

7 Lay out plates.

8 When you are ready, remove FSW-agar mix from 32 °C bath and allow to cool 1-2 degreesCritical: the mixture will start to solidify at 27-28 °C.

9 Add cell suspension to each tube, mix gently, then pour into plates.

10 Let solidify to a loose consistency

11 Transfer to growth room. Single colonies should appear within 10 days (without selection).