Name: Laura, Beenle
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Goal: Transform Symbiodinium and O.Marina using Lonza

Protocol:

1. 1.5 mL of the symbiodinium (cell count: 1,092,000 cells/mL) was added to an eppendorf tube and spun down at 900g for 3 minutes. 3.5 mL of the o. Marina (cell count: 420,000 cells/mL) was added to a 50mL falcon tube and spun down at 1000g for 3 minutes.
2. The supernatant was removed from both tubes and 98 µL of SG and 22 µL of the supplement were added to EACH of the tubes and were swirled to resuspend the pellet.
3. 40 µL aliquots were made of each cell type (6 tubes total)
4. O. marina tubes
    a. 20 µL of Dino III RFP (ng/µL)
    b. 20 µL of Dino III GFP (ng/µL)
    c. 20 µL diH2O (for the blank)
5. S. microadriaticum tubes
    a. 40 µL of Dino III RFP (ng/µL)
    b. 20 µL of Dino III GFP (ng/µL)
    c. 20 µL diH2O (for the blank)
6. The Lonza machine was set for the correct pulse codes (DS-137 and DS-130) and 25 µL of each sample was added to the corresponding cuvette well
7. After shocking, 80 µL of media (ASP8A for s. microadriaticum and F/2 for o. marina) was added to the cuvette well and pipetted up and down three times. The contents of the cuvette were then transferred to the corresponding well in a 24-well plate.
8. The 24-well plates were placed in artificial light (hub) for incubation.

Notes:

- On the 24-well plate, A stands for pulse code DS-137 and B stands for pulse code DS-130
- The 24-well plate on top contains transformed O. marina and the one on the bottom contains transformed S. microadriaticum