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Version 1

♦ NEPA electroporation of Emiliania huxleyi cells V.1

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Protocol status: Working

We use this protocol in our group for introducing proteins and DNA into E. huxleyi cells

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Abstract

We used the NEPA electroportor to transform Emiliania huxleyi cells.

We were able to establish that this method can be used to introduce proteins into the cells.

Troubleshooting



- 1 Collect exponentially growing E. huxleyi cells by centrifugation at 3000 g for 3 min, room temp.
- 2 Wash the cells once and resuspended with 384mM Sorbitol to a final concentration of ~108 cells/ml.
- 3 Mix 150 ul of cells with 1–10 mg of (linearized) plasmid and transfer to an electroporation cuvette with 0.2 cm gap.
- 4 Electroporate with optimal conditions – we used 7 poring pulses of 250V, followed by transfer 10 +/- transfer pulses.
- 5 After electroporation, transfer IMMIDIATELY into 4 mL of fresh media and incubate to allow recovery in nonselective medium in the growth room, low light for 16-20 h.
- 6 Apply selection and cross your fingers...