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Non-invasive DNA transfection of *B. glabrata* using polyethylenimine (PEI)

Chemicals

- Ampicillin (Amp)
- 0.2 mg/ml of plasmid DNA (pDNA)
- PEI transfection reagent
- Sterile distilled water

Protocols

Day 1: Treat snail(s) with 100 mg/ml Amp in distilled water (DW) overnight

Day 2: DNA/PEI complex preparation (N/P ratio* = 6) transfection (Knight *et al.,* 2011)

- Dilute one microgram of plasmid DNA in 0.5 ml of nuclease free water and vortex briefly. Keep the tube at room temperature (RT)
- Dilute *0.78 ml*** of PEI solution in 0.5 ml of nuclease free water and vortex briefly
- Add diluted PEI solution in to pDNA solution slowly and vortex. Let the complex forming at RT for 15 min
- Transfection: transfer the clean-dry snail from day 1 into the DNA/PEI complex mixture and let it culture for at least 24 hours
- Monitor the successful of transfection by several assay *e.g.* PCR with specific primer to the plasmid DNA or your gene of interest.

Note: *This protocol is for 1 snail transfection (1 snail per 1 ml final DNA/PEI mixture).*

* N/P ratio is a measure of the ionic balance within the complexes and is defined as the number of nitrogen residues of PEI per nucleic acid phosphate. For the **parasite transfection**, the suggested N/P ratio is between 8-11 (Liang S. Knight M and Jolly, 2013, Liang et al., 2014).

*** This volume would be changed according to the various N/P ratios*

References:

1. Knight M, Miller A, Lui Y, Scaria P, Woodle M, Ittiprasert W. Polyethyleneimine (PEI) mediated siRNA gene silencing in the *Schistosoma mansoni* snail host, *Biomphalaria glabrata*. PLoS Negl Trop Dis 2011: 5(7): e1212.
2. Liang S, Knight M, Jolly ER. Polyethyleneimine mediated DNA transfection in schistosome parasites and regulation of the WNT signaling pathway by a dominant-negative SmMef 2. PLoS Negl Trop Dis 2013: 7(7): e2332.
3. Liang S, Varrecchia M, Ishida K, Jolly ER. Evaluation of schistosome promoter expression for transgenesis and genetic analysis. PLoS One 2014: 9(5): e98302.

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