

The Liu Laboratory protocol — OCP-E.coli
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1. Transformed BL21 cells were grown overnight in TB medium at 37° C in the presence of three antibiotics (ampicillin (100 ug/mL, chloramphenicol (10 ug/mL) and streptomycin (10 ug/mL) ;
2. Dilute the culture for 100 times in TB, then cells were grown at 37° C for 8 h (OD₆₀₀ is about 0.6);
3. Then add L-arabinose (stock, -20° C) into the culture, the final concentration is 0.02%, the culture was grown at 37° C overnight;
4. In the next morning, dilute the culture 10 times using TB medium with 0.02% L-arabinose and three antibiotics; then put the culture into shaker, 200 rpm, 37° C, 8 h, OD₆₀₀ is about 1.0;
5. Add IPTG (stock, in -20° C) into the culture, the final concentration is 0.2 mM, incubate the culture at 28° C overnight, collect the cells in the next morning, store at -80° C.

Terrific Broth (TB medium)

Reagent	Quantity	Final Concentration
Yeast extract	24 g	24 g/L
Tryptone	20 g	20 g/L
Glycerol	4 mL	4 mL/L
Phosphate buffer (0.17 M KH ₂ PO ₄ , 0.72 M K ₂ HPO ₄)	100 mL	0.017 M KH ₂ PO ₄ 0.072 M K ₂ HPO ₄

Add 900 mL of deionized water to 24 g of yeast extract, 20 g of tryptone (if Sigma TB-Novagen premix (71754) is used, measure 47.6 g power and omit phosphate buffer), and 4 mL of glycerol. Shake or stir until the solutes have dissolved and sterilize by autoclave for 20 min at 15 psi (1.05 kg/cm²). Allow the solution to cool to ~60° C and add 100 mL of sterile phosphate buffer. Store TB at room temperature, it will keep for at least 1 yr.