

1. Materials that you received.

#RF-6: BL21(DE3) RFzero-iy  
Genotype: *prfA::zeo*, BAC8(cat), pidoTyrRS-MJR1-gent(pACYC)  
Antibiotics: Cm 17 mg/L, Gent 7 mg/L, Zeo 25 mg/L  
3-bromo/chloro/iodotyrosine: 0.1mg/ml

2. Medium used

The stub medium (1 mL) contains agar (1.5mg), NaCl (1mg), Yeast Extracts (0.25mg), Tryptone (0.5mg), and 3-bromotyrosine (0.1 mg).

The shipped stub was inoculated with a culture incubated in LB containing 0.1 mg/mL bromotyrosine, Cm 17 mg/L, Gent 7 mg/L, Zeo 25 mg/L.

3. How to supplement 3-bromo/chloro/iodotyrosine in the media

- (6) 3-halogenated tyrosines can be dissolved in 0.2N HCl at a conc. of 10mg/mL (100 × final conc.)
- (7) No sterilization is necessary for the solution.
- (8) Shelf life of the solution is a month at 4 degrees.
- (9) Add to the growth media and then neutralize with 2N NaOH just before the media preparation.
- (10) Antibiotics are not necessary to maintain strains, and may be used to avoid contamination by other lab bacteria.