

Auto-Induction

“Magic” Media

Making ZY Media

1. 928mL of ZY Media
 1. 10g Tryptone
 2. 5g Yeast
2. Autoclave ZY Media.

Adding Ingredients After Autoclaving

3. 50mL of 20xP
 4. 20mL 50x5052 (500 mLs)
 1. 125 mL Glycerol
 2. 12.5 g Glucose
 3. 50 g α -lactose
 5. 2mL 1M MgSO₄
 6. 0.2mL 1000x Trace Elements

 1. Add the remaining ingredients: Antibiotic and “glycerol stock”.
- Example:**
2. Add antibiotic (ampicillin for hsp27 R136W) for a final concentration of 1g/L
 3. Add glycerol stock, 1mL for 1L of media.
 4. Shake at 220rpm for 24 hours.
 5. Centrifuge at 6000xg for 30 minutes at 4°C.
 6. Resuspend in 20mL of Lysis Buffer
 7. Add a pinch of lysozyme and put in hoola shaker for 30 minutes at 4°C.
 8. Freeze/thaw 3X
 9. Continue with purification protocol.