

XL10-Gold Bacteria Transformation

1. Pre-chill 14 mL Falcon tubes on ice
2. Pre-warm NZY broth to 42°C in water bath (500 µL per transformation).
Always work beside a flame when opening NZY
3. Pre-warm LB/amp plates in 37°C incubator
4. Thaw bacterial cells on ice
5. Add 45 µL of cells to pre-chilled tubes.
6. Re-freeze any unused cells
7. Add 2 µL β-MercaptoEthanol (green top in XL10 box)
8. Incubate 10 min on ice while swirling every 2 min
9. Add 2 µL SDM DNA
10. Incubate cells on ice for 30 min
11. Heat shock cells in 42°C water bath for 30 sec without shaking
12. Place cells on ice for 2 min
13. Add 500 µL of pre-warmed NZY to tubes
14. Incubate tubes in 37°C shaker for 1hr at 225-275 rpm
15. Plate entire 550 µL cells on pre-warmed LB/amp plates. Always work beside a flame when plating cells
16. Spread with glass rod and turntable. Always work beside a flame when spreading cells
17. Let plates dry at room temperature
18. Incubate plates in 37°C incubator upside down for <16 hrs
19. Remove plates and parafilm.
20. Store at 4°C