



GS selection.

CleanCut GS CHO

Materials:

CDForti CHO medium (Gibco, Cat# A11483-01)

Anti-Clumping Agent (ACA) (Gibco, Cat# 01-0057AE)

Option 1: initial 2×10^6 cells

Day 2 after nucleofection: check viability. Viability should be 90% or higher.

Day 2: transfer cells into T-25 low attachment (2×10^6 cells in 5ml). Speed 90rpm, 8%CO₂ in Glutamine free medium.

Day 5-6: When cells reach 9×10^6 cells with good viability, transfer cells to unbaffled E-125 (0.3×10^6 cell/ml) in 30 ml medium. Shaker speed 125rpm, 8%CO₂ in Glutamine free medium. GS Selection: 14 days. Change medium every 2days. . Re-seed cells with 0.3×10^6 cells/ml every two days during Glutamine selection.

Option 2: initial 8×10^6 cells

Day 2 after nucleofection: check viability. Viability should be 90% or higher.

Day 2: (if viability is good) transfer cells to unbaffled E-125 (0.3×10^6 cell/ml) in 30 ml medium. Shaker speed 125rpm, 8%CO₂ in Glutamine free medium. GS Selection: 14 days. Change medium every 2 days. Re-seed cells with 0.3×10^6 cells/ml every two days during Glutamine selection.