Strains of E coli to use

9-7-87 TO Do

Strain of E coli To Use

1. To make cDNA library
To make plasmid DNA preps for transfection of mammalian cells

1) Use DH1. Prepare competent cells by Okayama method
0.3–1.0 X 10^9 Transformation
2) Prob OK to buy SCS1 (1X10^9) or use DH1 (our prep)

$\boxed{\text{recA}^-}$ endA$^-$ R$_K^-$ EcoP1$^-$
$\boxed{\text{probably mcrA}^+\text{, mcrB}^+\text{?}}$

2) To transform E coli with DNA that has replicated in mammalian cells

1) Don't have a recA$^-$ endA$^-$ mcrA$^-$ mcrB$^-$ R$_K^-$ E coli
a) check genotype of E coli GM 2163 · recA$^+$ or $^-$?

2) Use either

1) E coli ER 1451 (= JM 107 with mcrB$^-$ mutation)
Call NEB for this strain
= $\boxed{\text{recA}^+}$ endA$^-$ R$_K^-$ EcoP1$^-$ mcrA$^-$ mcrB$^-$

Derived from DH-1.
Don't grow plasmids long in the E coli better to shift to recA$^-$ DH1 etc
OK to shift to DH-1 after DNA has replicated (overnight growth) in ER 1451
Make Gradient DNA prep & transform DH1

Use for brief DNA replication (this or maximum overnight)
Don't Use for ~~DNA~~ extensive DNA replication