

# Transposition Mutagenesis Flow Chart

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## Step 1: Conjugation

What	Why
Resuspend doner cells by vortexing	mix with nutrient broth
Vortex recipient briefly	
Put 500 $\mu$ L doner and 500 $\mu$ L recipient in mating tube and vortexing briefly	mate two strains
Incubate in 37°C for 60 min (with balance)	start mating
Label information on the plate (name, day, medium, solution, dilution)	
Add 900 $\mu$ L saline to 7 tube labeled with $10^{-1} \sim 10^{-7}$	prepare to dilute
Add 100 $\mu$ L recipient to $10^{-1}$ , vortex	dilute
Repeat until $10^{-7}$	
Put 100 $\mu$ L last three tube onto nutrient agar plates and spread in circular fasion	control recipient group (without mating)
Put 100 $\mu$ L $10^0$	