Suggested Transformation Protocol:

1. Thaw cells on wet ice after removing from -70°C
2. Mix cells gently by lightly flicking the tube. Aliquot ~50µl of cells into a chilled 1.5ml centrifuge tube(s). Unused cells may be refrozen, but a small drop in efficiency may result. For optimal recovery, refreeze cells in a dry ice/ ethanol bath prior to -70°C storage.
3. Add DNA solution (≤5µl per 50µl cells) to cell suspension and gently swirl tube(s) for a few seconds to mix. If a control is desired, repeat this step with 2µl of the provided pUC19 (50 pg/µl) in a separate tube.
4. Incubate on ice for 30 minutes.
5. Place tube(s) in 42°C water bath for ~30 to 45 seconds without shaking.
6. Place tube(s) again on ice for ~5 minutes.
7. Dilute transformation reaction(s) to 1ml by addition of 950µl SOC medium provided.
8. Shake tube(s) ~200 rpm for 60 minutes at 37°C.
9. Plate by spreading 5-200µl of cell transformation mixture on LB agar plates containing appropriate antibiotic and incubate overnight at 37°C.

When performing the pUC19 control transformation, plate 10µl of the transformation mixture on a LB agar plate containing 100µg/ml ampicillin. To facilitate cell spreading, place a pool of SOC (100µl) onto surface of plate prior to addition of transformation mixture.

Transformation Efficiency Calculation for Control DNA

$$\text{Transformation Efficiency (cfu/µg pUC19 DNA)} = \frac{\text{\# colonies (colony forming units)}}{\text{pg pUC19 transformed}} \times \frac{10^6 \text{ pg}}{\text{ug}} \times \frac{\text{Final volume (µl) of transformation mix}}{\text{Volume plated (µl)}}$$

For example:
If 40 colonies were obtained after transforming 100pg of pUC19 and plating 10µl of the final 1ml transformation mixture, the calculated transformation efficiency would be:

$$\frac{400 \text{ cfu}}{100\text{pg pUC19}} = \frac{10^6 \text{ pg}}{\text{ug}} \times \frac{1000\text{ul}}{10\text{ul}} = 4 \times 10^8 \text{ cfu/ug pUC19}$$