Transformation Considerations

Competent Cells

The competence of a microorganism is dependent on its ability to uptake recombinant DNA and survive the introduction of foreign DNA into the cell. Different organisms vary in these capacities, however the basic principles of introduction are the same. Modifications to the cell membrane/wall of microorganisms must occur often, using either chemical modification or electric shock. After this damage, cells are recovered and calculated for their "uptake" efficiency—measured as colony forming units per microgram of DNA (cfu/µg). Some common transformation efficiencies are listed below in Table 1.

Table 1 - Transformation efficiencies (cfu/µg)

	Chemically Competent	Electrocompetent
E. coli	1.0×10^6 to 5.0×10^9	1.0×10^8 to 2.0×10^{10}
S. cerevisiae	1.0×10^3 to 2.2×10^7	1.0×10^5 to 1.0×10^7
S. pombe	1.0×10^3 to 1.0×10^6	1.0×10^5 to 1.0×10^6
P. pastoris	1.0×10^2 to 1.0×10^5	1.0×10^4 to 1.0×10^5

Transformation Method

Invitrogen offers chemically competent and electrocompetent *E. coli*. Chemically competent *E. coli* have a fragile cell wall which make cells prepared in this manner incompatible with electrocompetent transformation methods where a high-energy field is applied to the cells/DNA mixture. Likewise, Invitrogen's electrocompetent *E. coli* are not transformable with any heat-shock transformation technique.

Rapid Transformation Procedure for Use with TOPO® Vectors

Recommended only for transformations using ampicillin selection.

1. Add 4 µl of the TOPO® Cloning reaction to one vial of One Shot® Chemically Competent *E. coli* and mix gently.
2. Incubate on ice for 5 minutes.
3. Spread 50 µl of cells on a pre-warmed LB plate (containing ampicillin and X-gal) and incubate overnight at 37˚C.