

CLONING AND EXPRESSION

Transformation

Competent Cells in Convenient Formats

Sigma offers several *Escherichia coli* strains as chemically competent and electrocompetent cells in ready-to-use 50 μ l single tube aliquots or Uni-packs, 8 well strips, and standard aliquots. Uni-packs offer convenience by allowing you to thaw only the cells you need and reduces the number of pipetting steps. The 8 well strips provide a high throughput format for performing bacterial transformations. The cells are made chemically competent or electrocompetent using optimized procedures specific to each strain and are offered at transformation efficiencies of 10^6 to 10^{10} cfu/ μ g of pUC plasmid DNA for routine subcloning and recombinant protein expression to more sensitive applications.

Competent Cell Strain Selection Table

Feature	GC5™	Thunderbolt™				BL21	BL21	BL21	
		GC10™	GC10™	JM109	HB101		(DE3)	(DE3)	pLysS
Comparable to DH5 α ™	√								
Comparable to NovaBlue and XL1 Blue				√					
Comparable to DH10B™		√	√						
Strain Background	K12	K12	K12	K12	K12xB	B	B	B	B
Transformation Efficiency (cfu/ μ g pUC plasmid DNA)	10^9	10^9	10^{10}	10^8	10^8	10^6	10^6	10^6	10^6
Cloning Strain	√	√	√	√	√				
Expression Strain						√	√	√	√
Blue/White Selection (with appropriate construct)	√	√	√	√					
T1 bacteriophage resistant	√	√	√						
T7 promoter systems							√	√	√
F' episome for single strand rescue				√					
Ideal for cloning methylated DNA		√	√		√				
<i>lac</i> ^P (higher level of <i>lac</i> repressor)				√					

NEW GC5™ Competent Cells

GC5 chemically competent cells are comparable to the popular DH5 α ™ strain and carry *recA1* and *endA1* mutations that aid in plasmid stability and improved quality of prepared plasmid DNA. GC5 Competent Cells are offered at a high efficiency grade for subcloning and generation of cDNA libraries. The transformation efficiency of GC5 Uni-packs (50 μ l single use aliquots) and Standard Aliquots is 1×10^9 cfu/ μ g pUC19 plasmid DNA. pUC19 Control DNA is provided.

Product Code	Description	Size
G 3169	GC5™ Competent Cells, Uni-Pack	10 x 50 μ l 20 x 50 μ l
G 3044	GC5™ Competent Cells, Standard Aliquots	5 x 200 μ l

NEW GC10™ Competent Cells

GC10 chemically competent cells are comparable to the popular DH10B™ strain and carry *recA1* and *endA1* mutations that aid in plasmid stability and improved quality of prepared plasmid DNA. GC10 Competent Cells are offered at a high efficiency grade for subcloning and generation of cDNA libraries. The transformation efficiency of GC10 Uni-packs (50 μ l single use aliquots) and Standard Aliquots is 1×10^9 cfu/ μ g pUC19 plasmid DNA. pUC19 Control DNA is provided.

Product Code	Description	Unit
G 2919	GC10™ Competent Cells, Uni-Pack	10 x 50 μ l 20 x 50 μ l
G 2794	GC10™ Competent Cells, Standard Aliquots	5 x 200 μ l

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Genotype: F- Φ 80/*lacZ* Δ M15 Δ (*lacZYA-argF*)U169 *endA1 recA1 relA1 gyrA96 hsdR17* (*r_k⁻*, *m_k⁺*) *phoA supE44 thi-1* λ -T1R

Features & Benefits

- Comparable to DH5 α ™
- Suitable for larger plasmids
- Blue/white color selection
- T1 bacteriophage resistant

Genotype: F- *mcrA* Δ (*mrr-hsdRMS-mcrBC*) Φ 80/*lacZ* Δ M15 Δ *lacX74 endA1 recA1* Δ (*ara, leu*)7697 *araD139 galU galK nupG rpsL* λ -T1R

Features & Benefits

- Comparable to DH10B™
- Suitable for methylated DNA
- Blue/white color selection
- T1 bacteriophage resistant
- Useful for generating cDNA libraries and genebanks

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Genotype: F- *mcrA* Δ (*mrr-hsdRMS-mcrBC*) Φ 80d*lacZ* Δ M15 Δ *lacX74* *endA1* *recA1* Δ (*ara, leu*)7697 *araD139 galU galK nupG rpsL* λ -T1R

Features & Benefits

- Same benefits as GC10
- Electrocompetent format
- Highest efficiencies

Genotype: F- *traD36 proA*⁺*B*⁺ *lacI*^q Δ (*lacZ*)M15/ Δ (*lac-proAB*) *glnV44 e14*⁻ *gyrA96 recA1 relA1 endA1 thi hsdR17*

Features & Benefits

- Comparable to NovaBlue and X11 Blue
- Blue/white color selection
- Routine sub-cloning
- Single-strand propagation from M13 or phagemid vectors
- Higher levels of *lac* repressor

Genotype: F- *mcrB mrr hsdS20*(*r_B⁻ m_B⁻*) *recA13 leu ara-14 proA2lacY1 galK2 xyl-5 mtl-1 rpsL20*(*Sm*^r) *supE44* λ ⁻

Features & Benefits

- Methyl restriction deficient
- Useful for cloning genomic DNA
- Routine sub-cloning
- Suitable for plasmids not used for blue/white color selection

Genotype: F- *ompT hsdS_B*(*r_B⁻ m_B⁻*) *gal dcm*

Features & Benefits

- Higher levels of protein expression
- Increased stability of recombinant protein
- (DE3) strains for expression from vectors utilizing the T7 promoter
- Tighter control of "leaky" expression with pLysS and pLysE strains

NEW Thunderbolt™ GC10™ ElectroCompetent Cells

Thunderbolt GC10 cells have the same features as the GC10 chemically competent cells described on previous page and are comparable to the popular DH510B™ strain. Thunderbolt GC10 ElectroCompetent Cells are supplied at the highest efficiency available for subcloning and generation of cDNA libraries. The transformation efficiency of Thunderbolt GC10 ElectroCompetent Cells is 1×10^{10} cfu/ μ g pUC19 plasmid DNA. Two different formats are offered for compatibility with your particular equipment. pUC19 Control DNA is provided.

Product Code	Description	Unit
T 7699	Thunderbolt™ GC10™ ElectroCompetent Cells	5 x 80 μ l 5 x 100 μ l

NEW JM109 Competent Cells

JM109 is a cloning strain for the preparation of high quality plasmid DNA. JM109, a K strain bacterium, carries *recA1* and *endA1* mutations that aid in plasmid stability and improved quality of prepared plasmid DNA. JM109 Competent Cell Uni-packs are chemically competent cells provided in 50 μ l ready-to-use aliquots while JM109 Competent Cell 8 Well Strips are provided with 4 strips of tubes containing 240 μ l aliquots (20 μ l reaction size, 12 reactions per tube, 96 reactions per strip). Both formats are provided at an efficiency of 1×10^8 cfu/ μ g pUC18 plasmid DNA. SOC Medium and pUC18 Control Plasmid are provided.

Product Code	Description	Unit
C 4114	JM109 Competent Cells, Uni-pack	11 rxns
C 9239	JM109 Competent Cells, 8 Well Strips	384 rxns

NEW HB101 Competent Cells

HB101 is a cloning strain for the preparation of plasmid DNA and general recombinant DNA cloning experiments. HB101 is a hybrid K12 x B strain bacterium, carrying a *recA13* mutation and *hsdS20* (*r_B⁻ m_B⁻*) restriction minus genotype for plasmid and insert stability. HB101 Competent Cell Uni-packs are chemically competent cells at an efficiency of 1×10^8 cfu/ μ g pUC18 plasmid DNA provided in 50 μ l ready-to-use aliquots. SOC Medium and pUC18 Control Plasmid are provided.

Product Code	Description	Unit
C 5489	HB101 Competent Cells, Uni-pack	11 rxns

BL21 Competent Cells

BL21 is the strain of choice for high-level gene expression and production of recombinant proteins in bacterial systems. BL21 strains lack the *lon* and *ompT* proteases thus promoting stability of recombinant proteins. Strains designated as "DE3" carry a copy of the T7 RNA polymerase under control of the IPTG inducible *lacUV5* promoter, and as a result, are ideal for controlled expression of T7 promoter driven constructs. Tighter control is provided by strains carrying the pLysS or pLysE plasmids encoding T7 lysozyme, a natural inhibitor of T7 RNA polymerase, which reduces background levels of polymerase activity in uninduced cells. When the recombinant protein is toxic to the cell, the pLysE host is ideal because it produces higher levels of the inhibitor providing the most stringent control in T7 promoter based systems. BL21 Competent Cell Uni-packs are chemically competent cells at a value efficiency of 1×10^6 cfu/ μ g pUC18 plasmid DNA provided in 50 μ l ready-to-use aliquots. SOC Medium and pUC18 Control Plasmid are provided.

Product Code	Description	Unit
B 8808	BL21 Competent Cells, Uni-pack	11 rxns
B 8683	BL21 (DE3) Competent Cells, Uni-pack	11 rxns
B 8933	BL21 (DE3) pLysS Competent Cells, Uni-pack	11 rxns
B 9058	BL21 (DE3) pLysE Competent Cells, Uni-pack	11 rxns

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