RBC
It's the Joy of Discovery

www.real-biotech.com

Real Biotech NEWS

HOT Products

- **HIT Competent Cells™ (Non-Heat Shock Transformation)**
- **RBC T&A Cloning Kit**

NEW Products NEW!

- **HIT Competent Cells™- DH5a Bravo**
  High efficiency as electrocompetent cells. No expensive equipment required.
- **HIT Competent Cells™- DH10B**
  For propagation of large insert DNA library clones. This strain is resistant to Streptomycin.
- **HIT Competent Cells™- GM2163**
  For propagation of plasmid or cloned DNA to be cut with Dam or Dcm-sensitive restriction enzymes. This strain is resistant to Chloramphenicol.

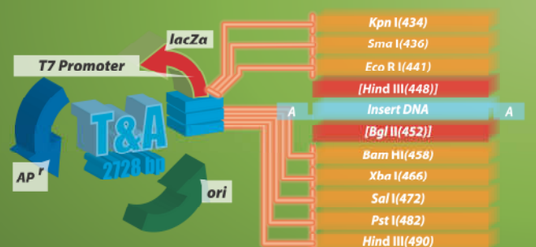
RBC Cloning Systems

RBC Rapid Ligation Kit

Cat.No. RC011 (100 reactions)
T4 DNA Ligase: (3 U/μl):100 μl
10X Ligation Buffer A: 200 μl
10X Ligation Buffer B: 200 μl

Store at -20°C

Map and Sequence Reference Points of the T&A Cloning Vector



Multiple Cloning Site	434	to	490
lacZα gene	511	to	149
AP^r gene	2528	to	1671
T7 promoter	402	to	439
M13 universal primer	359	to	375
M13 reverse primer	528	to	507

RBC Cloning Systems

RBC T&A Cloning Kit

Cat.No. RC001 (20 reactions)
T&A Cloning Vector (25 ng/μl): 40 μl
Control Insert DNA (10 ng/μl): 10 μl
T4 DNA Ligase (3 U/μl): 20 μl
10X Ligation Buffer A: 100 μl
10X Ligation Buffer B: 100 μl
Forward Primer(M13-F) (10 μM): 50 μl
Reverse Primer(M13-R) (10 μM): 50 μl

Store at -20°C

Cat.No. RC013 (20 reactions)
T&A Cloning Vector (25 ng/μl): 40 μl

Application

- Accepts terminal 3'-dA nucleotide overhang PCR products
- Transform ligation product (purified/unpurified) into HIT Competent Cells™
- LacZ complementation for Blue/White screening
- Ampicillin marker for antibiotic selection
- Universal primer for easy transformation screening

6 Minutes Cloning and Transformation

6 Minutes

- RBC T&A Cloning Kit (RC001)
- Purified DNA
- 22°C 5- 15 Minutes
- Ligated Vector with Insert
- <5% of total volume of competent cells
- HIT Competent Cells™
- 1 Minute
- Plate HIT with RBC Glass Beads (RG001)

- dA-tailed DNA (RT001-RealTaq DNA Polymerase) PCR Product Sticky
- Purify HiYield Gel/PCR Extraction Kit (YDF100)

- Agarose Gel Electrophoresis (check ligation)
- Purify/Inactive at 65°C, 10 Minutes HiYield Gel/PCR Extraction Kit (YDF100)

HIT™ Competent Cells
Real Biotech Corporation

Non-Heat Shock Transformation

HIT Competent Cells™

Advantage- Just Add Ice!
Compared To Traditional Commercial Competent Cells

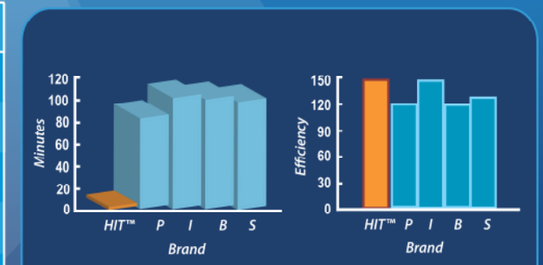
- Revolutionary 1 minute transformation
- -70°C monitoring of competent cell shipments
- True 1 step/1 tube process
- Guaranteed efficiency up to 10^9 cfu/μg plasmid
- Each batch efficiency checked during production
- Economic pricing for regular and high efficiency cells
- Ideal for bench users to automated cloning projects
- Worldwide patent protection

HIT Competent Cells™ Protocol Comparison Table

Competent Cell Types	HIT Competent Cells™	Traditional Chemically Competent Cells	Electro-Competent Cells
Time	1 minute	1.5~2.5 hours	1.5~2.5 hours
Protocol steps	One step No tube change/SOC	Multiple steps Requires tube change/ SOC	Multiple steps Requires tube change/SOC
Authentic representation	High	Requires recovery step	Requires recovery step
Transformation Efficiency	10^7~10^9 / μg pUC19	10^7~10^9 / μg pUC19	10^{11} / μg pUC19
Stability of refrozen cells	High	Low	Low
Stability of vortex	Stable	Unknown	Unknown

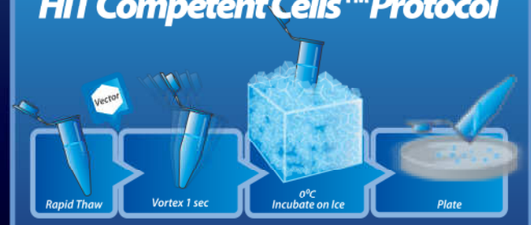
HIT Competent Cells™- DH5a Bravo, is as high efficiency as electrocompetent cells. No expensive equipment required!

Worldwide First: 1 Minute Transformation to High Efficiency



Efficiency Levels
HIT Competent Cells™ transformation efficiency reaches 10^7~10^9 transformation / μg pUC19 plasmid DNA.

HIT Competent Cells™ Protocol



Prepare ice bucket, 37°C plating beads and selective plates. Thaw competent cell vial with room temp. tap water or water bath for 10~20 seconds until 1/3 thawed.

Add DNA whose volume is less than 10% of volume of cells. Vortex 1 second.

Place on ice for 1~10 minutes.

Transfer onto 37°C dry selection plate media, spread using RBC plating beads.

Immediately incubate plate at 37°C (16~18 hours for HIT Competent Cells™-DH5a, 8~24 hours for HIT Competent Cells™-JM109). Observe growth of transformed colonies.

Worldwide publications describing the successful use of RBC products! Worldwide citations can be found on www.real-biotech.com!

Application Table

Cloning Applications	HIT™-DH5a	HIT™-JM109	HIT™-Blue	HIT™-21	HIT™-DH10B	HIT™-GM2163
Large Plasmids > 6 kb	Ideal	+	+	+	Yes	Yes
Subcloning	Ideal	Yes	Yes	+	Ideal	No
cDNA Library	Yes	Yes	Yes	+	Yes	+
Fast Growth	+	Ideal	+	Yes	+	+
Single Stranded DNA	+	Ideal	+	+	+	+
Toxic Protein Expression	No	No	No	No	No	No
Mutagenesis	Yes	+	+	No	Yes	+
Protein Expression	No	No	No	Ideal	+	No
Blue/White Screen	Yes	Yes	Ideal	No	Yes	No
DNA Unmethylation	No	No	No	No	No	Yes
Genomic DNA Cloning	No	No	No	No	Yes	No

+ Indicates that the strain can be used for the purpose, but may not yield the best result.

Genotypes Table

Genotypes	Applications	HIT™-DH5a	HIT™-JM109	HIT™-Blue	HIT™-21	HIT™-DH10B	HIT™-GM2163
		F- (80d lacZ M15) (lacZYA-argF)U169 hsdR17(r- m+) recA1 endA1 relA1 deoR	F' traD36 proA+ proB+ lacIq (lacZ)M15 (lac-proAB) hsdR17 recA1 endA1 relA1	hsdR17(rk- mk+), recA1, endA1, gyrA96, thi-1, supE44, relA1, lacI^q proAB lacIqZDM15Tn10(Tet^r)]	E.coli B, F-, dcm, ompT, hsdS(rB-mB-), gal (DE3)	F- endA1 recA1 galE15 galK16 nupG rpsL ΔlacX74 Φ80lacZΔM15 araD139 Δ(ara,leu)7697 mcrA Δ(mrr-hsdRMS-mcrBC) λ-	F- ara-14 leuB6 fhuA31 lacY1 tsx78 glnV44 galK2 galT22 mcrA dcm-6 hisG4 rfbD1 psL136 dam13::Tn9 xylA5 mtl-1 thi-1 mcrB1 hsdR2
endA1	Prevents plasmid degradation during extraction	Yes	Yes	Yes	No	Yes	No
recA1	Prevents DNA recombination	Yes	Yes	Yes	No	Yes	No
hsdR	Enhances transformation efficiency of selected PCR DNA strands and cDNA libraries	Yes	Yes	Yes	Yes	Yes	Yes
deoR	Enhances transformation efficiency of high MW plasmids and cosmids	Yes	No	No	No	Yes	No
LacZ M15	Inhibits LacZ gene expression for blue-white screen	Yes	Yes	Yes	No	Yes	No
Lon & ompT	Lon & ompT protease deficient and improves protein yield	No	No	No	Yes	No	No
rne131	Inhibits RNase E and improves mRNA stability	No	No	No	Yes	No	No
dam/dcm	Prevents DNA methylation	No	No	No	Yes / No	No	Yes
mcrA/mcrB	Prevents DNA methylated DNA from degradation	No	No	No	No	Yes	Yes

RBC Product Selection Guide

HIT™-DH5α

Strain: DH5α

HIT™-DH5α Value 10^8
(2 x 10^8 transformants/μg pUC19)
Cat. No. RH617
10 vials x 100 μl Competent Cells
Cat. No. RH617-J80
80 vials x 100 μl Competent Cells
Cat. No. RH617-96
96-Well x 50 μl Competent Cells

HIT™-DH5α High 10^8
(5 x 10^8 transformants/μg pUC19)
Cat. No. RH618
10 vials x 100 μl Competent Cells
Cat. No. RH618-J80
80 vials x 100 μl Competent Cells
Cat. No. RH618-96
96-Well x 50 μl Competent Cells

HIT™-DH5α Super 10^8
(2 x 10^9 transformants/μg pUC19)
Cat. No. RH619
10 vials x 100 μl Competent Cells
Cat. No. RH619-J80
80 vials x 100 μl Competent Cells
Cat. No. RH619-96
96-Well x 50 μl Competent Cells

NEW
HIT™-DH5α Bravo 10^9
(8 x 10^9 transformants/μg pUC19)
Cat. No. RH6110
10 vials x 100 μl Competent Cells
Cat. No. RH6110-J80
80 vials x 100 μl Competent Cells
Cat. No. RH6110-96
96-Well x 50 μl Competent Cells

Genotype:
F- (80d lacZ M15) (lacZYA-argF)U169 hsdR17(r - m +) recA1 endA1 relA1 deoR

Most popular cloning strain worldwide. Recommended for General Cloning, TA Cloning, Blue/White Screening, cDNA Library Construction.

HIT™-DH10B

NEW
Strain: DH10B

HIT™-DH10B Value 10^8
(1 x 10^8 transformants/μg pUC19)
Cat. No. RH517
10 vials x 100 μl Competent Cells
Cat. No. RH517-J80
80 vials x 100 μl Competent Cells
Cat. No. RH517-96
96-Well x 50 μl Competent Cells

HIT™-DH10B High 10^8
(2 x 10^8 transformants/μg pUC19)
Cat. No. RH518
10 vials x 100 μl Competent Cells
Cat. No. RH518-J80
80 vials x 100 μl Competent Cells
Cat. No. RH518-96
96-Well x 50 μl Competent Cells

80 vials of 100 μl Competent Cells

Genotype:
F- endA1 recA1 galE15 galK16 nupG rpsL ΔlacX74 Φ80lacZΔM15 araD139 Δ(ara,leu) 7697 mcrA Δ(mrr-hsdRMS-mcrBC) λ-T1R

For propagation of large insert DNA library clones.
This strain is resistant to Streptomycin.
Recommended for General Cloning, TA Cloning, Blue/White Screening, cDNA Library Construction.

HIT™-Blue

Strain: XL1-Blue

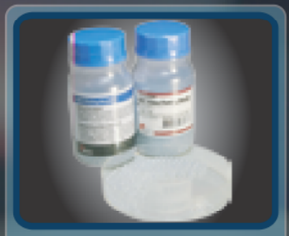
HIT™-Blue Value 10^8
(1 x 10^8 transformants/μg pUC19)
Cat. No. RH117
10 vials x 100 μl Competent Cells
Cat. No. RH117-J80
80 vials x 100 μl Competent Cells
Cat. No. RH117-96
96-Well x 50 μl Competent Cells

HIT™-Blue High 10^8
(5 x 10^8 transformants/μg pUC19)
Cat. No. RH118
10 vials x 100 μl Competent Cells
Cat. No. RH118-J80
80 vials x 100 μl Competent Cells
Cat. No. RH118-96
96-Well x 50 μl Competent Cells

HIT™-Blue Super 10^8
(2 x 10^9 transformants/μg pUC19)
Cat. No. RH119
10 vials x 100 μl Competent Cells
Cat. No. RH119-J80
80 vials x 100 μl Competent Cells
Cat. No. RH119-96
96-Well x 50 μl Competent Cells

Genotype:
recA1 endA1 gyrA96 thi-1 hsdR17 supE44 relA1 lac [F'proAB lacI^q ZΔM15 Tn10(Tetr)]

Most popular Blue/White screening strain. Recommended for General Cloning, Subcloning, and TA Cloning.



RBC Glass Plating Beads
Cat.No. RG001
1 x 100 g RBC Glass Plating Beads (Sterile)

HIT™-JM109

Strain: JM109

HIT™-JM109 Value 10^8
(1 x 10^8 transformants/μg pUC19)
Cat. No. RH717
10 vials x 100 μl Competent Cells
Cat. No. RH717-J80
80 vials x 100 μl Competent Cells
Cat. No. RH717-96
96-Well x 50 μl Competent Cells

HIT™-JM109 High 10^8
(3 x 10^8 transformants/μg pUC19)
Cat. No. RH718
10 vials x 100 μl Competent Cells
Cat. No. RH718-J80
80 vials x 100 μl Competent Cells
Cat. No. RH718-96
96-Well x 50 μl Competent Cells

Genotype:
e14$^-$ (McrA$^-$) recA1 endA1 gyrA96 thi-1 hsdR17(r_K^- m_K^+) supE44 relA Δ (lac-proAB) [F' traD36 proAB lacI^q ZΔ M15]

Rapid growth!
Results in pickable colonies within 8-10 hours.
Recommended for General Cloning, TA Cloning, Blue/White Screening, Robotic Screening, Gene Library Construction.

HIT™-GM2163

NEW
Strain: GM2163

HIT™-GM2163 Value 10^8
(1 x 10^8 transformants/μg pUC19)
Cat. No. RH317
10 vials x 100 μl Competent Cells
Cat. No. RH317-J80
80 vials x 100 μl Competent Cells
Cat. No. RH317-96
96-Well x 50 μl Competent Cells

Genotype:
F-ara-14 leuB6 fhuA31 lacY1 tsx78 glnV44 galK2 galT22 mcrA dcm-6 hisG4 rfbD1 rpsL136 dam13::Tn9 xylA5 mtl-1 thi-1 mcrB1 hsdR2

For propagation of plasmid or cloned DNA to be cut with Dam or Dcm-sensitive restriction enzymes.
This strain is resistant to Chloramphenicol. Not suitable for blue/white screening.

HIT™-BL21

Strain: BL21 (DE3)

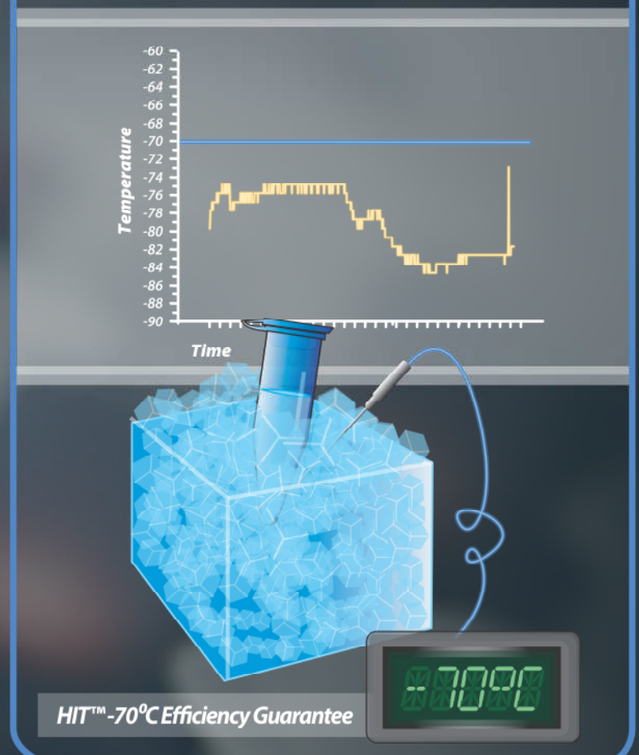
HIT™-BL21 Value 10^8
(3 x 10^7 transformants/μg pUC19)
Cat. No. RH217
5 vials x 100 μl Competent Cells
Cat. No. RH217-J40
40 vials x 100 μl Competent Cells
Cat. No. RH217-96
96-Well x 50 μl Competent Cells

Genotype:
E. coli B F- ompT dcm hsdSβ(rβ - mβ -) galλ (DE3)

Most popular strain worldwide for high level inducible protein expression. Recommended for Protein Expression.

Shipping Guarantee

International Shipping Temperature Report
Departure time: 06/17/03 11:16:32.0 (Departure)
Arrival time: 06/19/03 08:55:32.0 (Arrival)



HIT™-70°C Efficiency Guarantee

HIT Competent Cells™ QC

Efficiency test and α-complementation test

- Thawing HIT Competent Cells™ with RT water for 20 seconds until 1/3 thawed.
- Add 1μl (10^{-6} μg/μl) pUC19 (ice-bathed plasmid).
- Vortex 1 second.
- Place on ice for 5 minutes.
- Using RBC plating beads for plating on LB plate which is with 50 μg/ml Amp.
- Immediately incubate the plate at 37°C for 16 hours.

Ampicillin-resistance test

- Thawing HIT Competent Cells™ with RT water for 20 seconds until 1/3 thawed.
- Vortex 1 second.
- Plate 50 μl cells on LB plate which is with 50 μg/ml Amp.
- Immediately incubate the plate at 37°C for 16 hours.
- Plate the plate at RT for 3 days (Contamination test).

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Real Biotech Corporation is a leading R&D based life science solutions provider.
For more information on our extensive and innovative life science range, please visit our website www.real-biotech.com.

Ordering Information
To inquire product information or to place orders, please contact your local distributor. If you have questions about ordering or distribution process, please contact us. ▶ info@real-biotech.com

Technical Support
Our technical support department is staffed by experienced molecular biologists who are closely involved in the development of our products. They are committed to handling your technical inquiries quickly and efficiently. To ask technical questions, please contact us. ▶ support@real-biotech.com

Satisfaction Guarantee
We hope you could be completely satisfied with products and service provided by Real Biotech Corporation. If any product and service fails to meet your expectations, please feel free to contact us anytime! We are pleased to do our best to ensure your satisfaction. ▶ service@real-biotech.com