

Transform & Grow™ Bacterial Transformation Kit

*For rapid and user-friendly preparation of
transformation-competent bacterial cells*

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Application Manual

Revision # 2150-999-3C01

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50 Samples

Storage:

Ambient temperature (15–30°C)

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1. Introduction

Transformation is a common method of introducing foreign DNA into a bacterial host cell. An essential part of the DNA cloning process, bacterial transformation steps are required at many points during the progression of a molecular biology experiment. Transformation is used to introduce plasmid ligation reactions into bacteria for clone replication. Recombinant plasmids are transformed into different bacterial host cells for large scale replication and for maintenance and identification, to express proteins, to study mutagenized clones and to meet a wide variety of other research needs.

Bacterial transformation was first demonstrated by Mandel and Higa (1) who reported that bacteriophage DNA introduced into *E. coli* cells produced infectious phage centers. Cohen and colleagues subsequently demonstrated calcium-dependent *E. coli* transformation with plasmids carrying antibiotic resistance that resulted in the plasmid DNA being maintained in the cell as independently replicating episomes (2). Following bacterial cell transformation, the plasmid is partitioned to daughter cells during cell division.

Bacterial transformation methods generally fall into the category of either chemical transformation or electroporation. Both methods begin by treating bacterial cells with specific chemicals to prepare them for DNA uptake (make them “competent”) and to enhance the plasmid transfer through the bacterial cell wall.

Electroporation combines this chemical preparation of bacterial cells with the use of electric potential to introduce DNA into the cell. Electroporation provides a transformation efficiency up to 10^9 transformants per microgram plasmid DNA and is typically required for library screening to satisfy the need for increased clone representation. Electroporation is not a critical parameter for success when screening a ligation reaction for a clone containing insert DNA or transforming plasmid into a new host strain.

Many variations of the chemical transformation method have been developed in different laboratories with the intent of optimizing the transformation process. The methods may include specialized buffers, may incorporate heat shock or incubation on ice during the transformation process, or they may require growing the bacteria at a reduced temperature to increase transformation efficiency (3, 4). Chemical transformation methods are reported to provide a transformation efficiency up to 10^8 transformants per microgram plasmid DNA, but these results are often not reliably reproduced. The Qbiogene Transform & Grow™ Bacterial Transformation Kit has been developed to minimize the number of reagents, procedure steps and hands-on time required to reliably obtain consistent transformation efficiency.

The Transform & Grow™ Bacterial Transformation Kit provides a simple and efficient method for the preparation, transformation and storage of competent *E. coli* cells. The transformation technique, based on observations by Chung et al. (5), grows bacterial cells at 37°C, harvests them in mid-log growth and resuspends them in Transform-It™ Solution. This unique buffer both enables the cells to become immediately available for transformation and permits freezing the cells for future use without further preparation. The competent

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cells remain stable at -70°C to -80°C for at least 10 weeks. The Transform & Grow™ Bacterial Transformation Kit eliminates the requirement for heat shock and prepares competent cells within two and a half hours with only 30 minutes of hands-on manipulation. The kit contains the necessary reagents to prepare competent cells with a reliable and reproducible transformation efficiency of $2 - 8 \times 10^6$ transformants per μg of supercoiled plasmid DNA.

In addition to the unique Transform-It™ Solution and BIO 101® Systems SUPER-COMP Media, the Transform & Grow™ Bacterial Transformation Kit includes Roll & Grow™ Plating Beads to replace the traditional hockey stick and flame method for plating cells on solid growth media. The sterile Roll & Grow™ Plating Beads are added directly to the growth plate surface with the transformed bacterial cells, rolled back and forth to disperse the cells, and then discarded prior to incubation.

2. Kit Components and User Supplied Materials

2.1 Transform & Grow™ Bacterial Transformation Kit Components

SUPER-COMP Media	120 ml
Transform-It™ Solution*	6 ml
Roll & Grow™ Plating Beads	1 each
Short Protocol	1 each
User Manual	1 each
MSDS	1 each
Certificate of Analysis	1 each

**Transform-It™ Solution is stored at room temperature, but should be chilled on ice at least 15 minutes prior to use.*

2.2 User Supplied Materials

LB (Luria-Bertani) Broth or CIRCLEGROW® liquid culture media (see Related Products Section)

5 ml sterile culture tubes for growth of the overnight bacterial culture

50 ml or larger sterile culture tubes or flasks for growth of diluted overnight culture

1.5 or 2.0 ml microcentrifuge tubes for the transformation reaction

15 ml sterile aerated polypropylene culture tubes for cell recovery

Agar plates for bacterial culture

37°C environmental shaking incubator

Pipettmen and tips

Ice

Spectrophotometer

3. Important Considerations Before Use

3.1. Aseptic Technique

Practice aseptic technique when using the Transform & Grow™ Bacterial Transformation Kit.

- Minimize the introduction of contamination into the cell culture from dust, hair and clothing.
- Treat the benchtop work area prior to use with 10% bleach or 70% ethanol to remove potential contaminants.
- Use sterile microcentrifuge tubes, glassware, plates, pipettes and tips.
- Flame-sterilize open liquid containers before and after removing the cap.
- Aliquot reagents to prevent contaminating the master stock.

3.2 Culture Growth Conditions

Adequate aeration of the growing bacteria is critical. The media volume should be 25% or less of the culture container and the inoculated culture should be agitated to provide maximal air transfer to the media.

Harvesting bacterial cells during middle log phase (OD_{600} of 0.5-0.6) yields optimal transformation efficiency. Cells can be harvested during stationary phase, but transformation efficiency will be significantly reduced.

The Transform & Grow™ Bacterial Transformation Kit protocol allows for the use of 5 ml of the cell culture for determining the bacterial OD_{600} reading for harvest. Cell density may double within 10–15 minutes during the log growth phase.

3.3 Plasmid DNA

Transformation efficiency is dependent on plasmid size, concentration and degree of supercoiling, and may need to be titrated for maximal efficiency. In general, the transformation efficiency will plateau at approximately 500 nanograms of plasmid DNA. As little as 5 nanograms of plasmid will provide approximately $1 - 4 \times 10^4$ colonies with the Transform & Grow™ Bacterial Transformation Kit.

The transformation efficiency of ligated DNA is approximately 100-fold less than covalently closed supercoiled DNA. When transforming a ligation reaction, it is recommended that 1 μ l, 5 μ l and 10 μ l of a 25 μ l reaction are used to accommodate variability in the ligation efficiency.

3.4 Agar Plates

It is strongly recommended that growth plates used for plating the transformation reaction be pre-warmed to 37°C before use. Cold plates have been experimentally demonstrated to result in reduced transformation efficiency.

3.5 Transform-It™ Solution

The Transform-It™ Solution is stored at room temperature. This reagent requires chilling on ice before use but should be returned to room temperature for storage.

3.6 Incubation Time

The Transform & Grow™ Bacterial Transformation Kit permits flexible incubation times. Cells resuspended in Transform-It™ Solution may be incubated on ice for 15 – 60 minutes prior to the addition of DNA without significantly affecting the transformation efficiency.

3.7 Recommended Controls for All Transformations

- Negative Control: Competent bacteria without added plasmid should be plated on positive selection media to confirm the selection is functioning and that the bacteria used to prepare competent cells do not contain plasmid prior to use.
- Positive Control: Transform a preexisting plasmid at a known concentration to confirm cell competency, check positive selection and permit calculation of transformation efficiency.

3.8 Recommended Controls for Ligation Transformations

In addition to the controls suggested in Section 3.7, the following controls are strongly recommended for the transformation of ligation reactions:

- Untreated parent plasmid is used to indicate the transformation efficiency.
- Prepared (e.g. restriction digested, phosphatase treated, etc.) vector without DNA insert in a ligation reaction (lacking ligase) is used to indicate the potential background from undigested or self-ligating parent vector.
- Insert alone is transformed to indicate if the parent vector containing the insert was completely removed from the insert prior to ligation and transformation.

4. How to Calculate Transformation Efficiency

Transformation efficiency is calculated as Colony Forming Units (CFU) per microgram of DNA. Each CFU is the result of being transformed with at least one plasmid. However, if the amount of plasmid DNA is too high, a cell may become transformed with multiple plasmids. This is not often a problem when an existing plasmid is transformed among different strains when using nano- or picograms of DNA. It may become a problem during a ligation transformation if a high percentage of parent vector from the plasmid or the insert is present.

To Calculate Transformation Efficiency:

$$\left[\frac{\# \text{ of Colonies}}{\text{on Plate}} \right] \times \left[\frac{\text{Total Cell Volume } (\mu\text{l})}{\text{Volume Plated } (\mu\text{l})} \right] \times \left[\frac{1}{\mu\text{g transformed}} \right] = \text{CFU}/\mu\text{g}$$

Example: 100 μl competent cells containing 5 ng (0.005 μg) plasmid DNA was diluted into 900 μl SUPER-COMP Media for 37°C incubation. 100 μl was spread on the plate and 697 colonies were counted.

$$\left[697 \right] \times \left[\frac{1000 \mu\text{l}}{100 \mu\text{l}} \right] \times \left[\frac{1}{0.005 \mu\text{g}} \right] = 1.39 \times 10^6 \text{ transformations per } \mu\text{g DNA}$$

5. Safety Precautions

The Transform & Grow™ Bacterial Transformation Kit contains components that when in contact with human tissue or during inhalation may cause irritation. Wear personal protective equipment to prevent skin contact (e.g. gloves, lab coat, and eye protection) and prevent inhalation of reagent vapors and consumption of liquid during use. Consult the enclosed Material Safety Data Sheet for additional details.

6. Quick Protocol for Experienced Users

Preparation of Competent Cells:

1. Inoculate a single colony in 4 ml LB or CIRCLEGROW® Broth and grow overnight with shaking at 37°C.
2. Prepare a 1:50 dilution of the overnight culture in SUPER-COMP Media to provide sufficient volume for 1 ml for each transformation and 5 ml for the OD₆₀₀ determination.
3. Incubate at 37°C to an OD₆₀₀ of 0.5-0.6.
4. Transfer cells to a sterile centrifuge tube and place on ice for 15 minutes.
5. Centrifuge at 3000 x g at 4°C for 15 minutes. Pour off supernatant.
6. Resuspend cells in ice-cold Transform-It™ Solution (100 μl for each transformation reaction).
7. Chill cells on ice for 15 minutes.
8. Transfer 100 μl of cells per transformation reaction to prechilled (on ice) sterile microcentrifuge tubes.

Transformation of Plasmid DNA:

1. Add DNA to each 100 µl aliquot of chilled cells.
2. Incubate on ice for 10 minutes.
3. Transfer to room temperature for 10 minutes.
4. Return to ice for 10 minutes.
5. Transfer cells to a sterile aerated 15 ml culture tube containing 0.9 ml SUPER-COMP Media.
6. Incubate with moderate shaking at 37°C for 60 minutes.
7. Pre-warm selective agar plates at 37°C.
8. Plate 100 µl of culture using Roll & Grow™ Plating Beads.
9. Incubate plates inverted at 37°C overnight.

7. Detailed Protocol

7.1 Culture Preparation

1. Prepare single colonies for inoculation (Day 1)

Streak the bacterial strain that will be transformed onto a solid growth media to obtain single colonies. Use antibiotic selection if required. Incubate the bacterial plate inverted overnight at 37°C to obtain single colonies.

2. Prepare initial overnight culture (Day 2)

Using aseptic technique, transfer 4 ml LB or CIRCLEGROW® media to a sterile growth container. Inoculate the liquid media with a single bacterial colony from the Day 1 plate and grow overnight at 37°C in an environmental shaker incubator at ~200 rpm.

3. Grow a sufficient number of cells for transformation (Day 3)

Each transformation reaction requires a liquid volume of 1 ml of cells at an OD₆₀₀ of 0.5 - 0.6. The recommended total culture volume required includes an additional 5 ml of culture to permit checking the OD₆₀₀ reading during growth to determine the harvest point. See sections 3.7 and 3.8 for recommended transformation controls.

As shown in the following table, use 1 part overnight culture to 49 parts SUPER-COMP Media to prepare a final culture dilution of 1 in 50. For example, if 25 transformation reactions are required, a total culture volume of 30 ml is necessary. Combine 29.4 ml SUPER-COMP Media and 600 µl of the overnight culture. This will provide 5 ml of cell culture to check the OD₆₀₀ reading and 1 ml for each of the 25 transformation reactions.

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Component	Volume for:			
	5 Reactions	10 Reactions	15 Reactions	25 Reactions
SUPER-COMP Media	9.8 ml	14.7 ml	19.6 ml	29.4 ml
Initial overnight culture for 1:50 dilution	200 µl	300 µl	400 µl	600 µl

Incubate the cells at 37°C in an environmental shaking incubator at ~200 rpm until the cells reach a density at OD₆₀₀ of 0.5 -0.6

Note: *Harvesting the cells at OD₆₀₀ of 0.5 - 0.6 is recommended for optimal transformation efficiency. Different E. coli strains will vary in the time it takes to reach this point.*

7.2 Preparation of Competent Cells

1. Transfer the cells to a sterile centrifuge tube. Place on ice for 15 minutes.
2. Place the bottle of Transform-It™ Solution and the required number of empty, sterile microcentrifuge tubes on ice.
3. Pellet the cells by centrifugation at 3,000 x g at 4°C for 15 minutes. Discard supernatant.
4. Determine the volume of Transform-It™ Solution required to resuspend the pellet.
Each transformation reaction requires a liquid volume of 100 µl of resuspended cells. To calculate the volume of Transform-It™ Solution required (in milliliters), multiply the total number of reactions required (determined in Section 7.1, Step 3) by 0.1.

For example: If 10 transformations are required, 10 ml of culture will remain after removing 5 ml to determine the OD₆₀₀ (Section 7.1, Step 3). The cell pellet will therefore be resuspended in 1 ml of Transform-It™ Solution (10 ml x 0.1 ml).

5. Resuspend the pellet in the volume of ice-cold Transform-It™ Solution determined in Step 4.
6. Incubate the resuspended cells on ice for 15 minutes.

Note: *If transformation of DNA is to be performed the same day, resuspended cells can be left on ice at this step for up to 60 minutes without affecting transformation efficiency. If cells are to be frozen for future use, do not incubate longer than 15 minutes. Proceed with Step 7 and store cells at -70°C.*

7. While maintaining cells on ice, transfer 100 µl per transformation reaction to the chilled, sterile microcentrifuge tubes.

8. Proceed with the transformation protocol (Section 7.3) or store the competent cells at -70°C for future use.

Note: *Competent cells are stable at -70°C for at least 10 weeks with minimal decrease in transformation efficiency.*

7.3 Transformation of Plasmid DNA

Note: *Competent cells prepared immediately prior to use may be stored on ice for up to 60 minutes (45 minutes beyond the recommended 15 minutes) before adding DNA without loss of transformation efficiency (Section 7.2, Step 6).*

Note: *Frozen competent cells should be thawed on ice before proceeding to the next step.*

1. Prepare DNA for transfer to competent cells.

Note: *Refer to Sections 3.7 and 3.8 for appropriate transformation control reactions.*

Note: *The guidelines for DNA volume given below reflect typical experimental settings for transformation of ligations and supercoiled plasmids. It is only critical to accurately pipet a known quantity of DNA when measurements of transformation efficiency will be determined and compared to other readings or used as standards (see Section 3.7 for appropriate controls).*

For transformation of supercoiled plasmid DNA, prepare a dilution such that amounts of 1 ng, 5 ng and 10 ng may be added using volumes between 5 μl and 20 μl . Overloading the cells with plasmid is not recommended and can lead to a decrease in efficiency and/or result in a single cell taking up more than one plasmid. While a standard pUC-based vector may show increased efficiencies for up to 1000 ng transformed, the addition of more DNA in order to increase efficiency must be titrated for each plasmid/host combination.

For transformation of ligation reactions, volumes of 1 μl , 5 μl and 10 μl are recommended. In general, more DNA is required for efficient transformation of a ligation reaction containing open circular and/or nicked DNA than for supercoiled DNA.

2. Add plasmid DNA to each 100 μl aliquot of chilled competent cells.
3. Gently finger-flick tubes to mix and incubate on ice for 10 minutes.
4. Transfer tubes to room temperature for 10 minutes.
5. Place tubes back on ice and incubate for 10 minutes.
6. Add 0.9 ml SUPER-COMP Media to a sterile aerated 15 ml culture tube.

- Using aseptic technique, transfer the entire volume of transformed cells to the 0.9 ml SUPER-COMP Media and incubate at 37°C in an environmental shaker incubator at ~100 rpm for 60 minutes.

Note: *Incubation permits bacterial cell recovery and expression of the antibiotic resistance gene.*

Note: *Some plasmid/host combinations will provide 30% - 40% of the total possible number of transformants with no incubation step. However, optimal transformation efficiency is achieved after incubation for 60 minutes.*

- Pre-warm the appropriate number of selective agar plates at 37°C.
- Transfer 100 µl of the recovered cells onto the pre-warmed plates and evenly disperse them using Roll & Grow™ Plating Beads as follows:
 - Aseptically dispense 6 - 7 Roll & Grow™ Plating Beads onto each agar plate.
 - Replace the plate cover and gently roll the beads back and forth for approximately 30 seconds to disperse the cells. Multiple plates may be stacked and rolled simultaneously.
 - Remove the lid and roll beads from the plate into a biohazard waste receptacle.
- Replace the lid and incubate the plates inverted at 37°C overnight.

8. Common Questions

8.1 Does Transformation Efficiency Vary with the Volume of Competent Cells Produced?

The transformation efficiency of the Transform & Grow™ Bacterial Transformation Kit does not significantly fluctuate with the total volume of competent cells produced. Expected transformation efficiency normally varies between 2 - 8 x 10⁶ transformants per microgram DNA.

8.2 Does Transformation Efficiency Vary with Plasmid Size?

It is well known that there is an inverse relationship between transformation efficiency and plasmid size. For example, there can be a tenfold decrease in efficiency from a 3 kb plasmid to a 14 kb plasmid.

Transformation efficiency for larger plasmids may be increased by adding an increased amount of plasmid DNA (See Figure 1). A titration of plasmid DNA concentrations may be needed to find the optimal concentration for a specific plasmid. It has also been observed that lowering the antibiotic concentration in the growth plate may enhance cell survival with larger plasmids.

Plasmid Name	Plasmid Size	Concentration Range (ng)	Transformation Efficiency
pUC18	2686 bp	5 - 1000	$4.1 - 7.1 \times 10^6$
pBKRSV	4.4 kb	10 - 50	$3.0 - 5.4 \times 10^6$
pGEX4T	4.696 kb	5 - 10	$1.2 - 1.6 \times 10^6$
PlasmidMA	~6.5 kb	25	1.3×10^6
PlasmidMB	6 - 7 kb	25	1.4×10^6
pCMV-Luciferase	9 kb	25	$4.2 - 6.7 \times 10^6$
PlasmidPLRC	12.8 kb	300	1.5×10^6
CosmidMA	45 kb	1000 - 1500	$5.3 - 7.1 \times 10^5$
CosmidMB	45 kb	500 - 1000	$1.1 - 1.7 \times 10^6$
CosmidMC	45 kb	1000	1.8×10^5
CosmidMD	45 kb	1000	7.7×10^5

Figure 1: Relationship between plasmid size, DNA concentration, and transformation efficiency.

8.3 Do Plasmids Containing Different Antibiotic Resistance Genes Have Different Transformation Efficiencies?

Transformation efficiency is not dependant upon the selective marker carried by the plasmid used in the transformation procedure.

9. Troubleshooting

9.1 Why don't I have colonies on my supercoiled plasmid transformation plate?

DNA was not added to the transformation mix

Review DNA origin. Was plasmid or insert DNA in the original tube? Was it possibly mislabeled? Was it an old ligation tube that did not produce colonies before?

Ensure DNA was added to the transformation mix.

Ensure plasmid DNA was properly diluted to provide 1 - 10 nanograms per transformation.

Ensure the proper amount of DNA was added if relying on an OD₂₆₀ reading for concentration.

Analyze an aliquot of the DNA by gel electrophoresis to confirm concentration, size and integrity.

DNA was not stored correctly and has degraded

Store purified DNA at -20°C to prevent DNase digestion.

Repurify the DNA from bacteria.

Analyze an aliquot of the DNA by gel electrophoresis to confirm concentration, size and integrity. Retransform if the DNA does not appear completely degraded.

Cells were not made competent

Repeat the transformation with a positive control: Use a preexisting supercoiled plasmid at a known concentration to confirm cell competency. Calculate the transformation efficiency.

Ensure instructions in the Detailed Protocol (Section 7) were followed correctly.

Incorrect antibiotic selection was used in the plate

Review the known antibiotic resistance of the plasmid.

Confirm growth media plates contain the correct selection. Streak plates with a host cell containing a plasmid with the correct selection resistance for confirmation.

Confirm the plates were produced with the correct concentration of antibiotic (e.g. diluted correctly for final concentration).

Plates were not incubated at the optimal temperature for at least 24 hours

Incubating bacteria above approximately 39°C will prevent bacterial growth. Check the incubator temperature with a calibrated thermometer. If using a bulb thermometer, place it into water in a beaker that has been equilibrated to incubator temperature before reading the value.

Incubating bacteria below 37°C will slow the growth rate to require longer incubation. Determine the incubator temperature as described above. If the incubator was below temperature, continue the incubation at the correct temperature and/or repeat the transformation.

9.2 Why don't I have colonies on my ligation transformation plate?

Ligation efficiency influenced the transformation outcome

Analyze a portion of the ligation reaction by agarose gel electrophoresis to determine if insert has been ligated to vector.

Increase the amount of ligation transformed to compensate for potential low ligation efficiency.

Pellet the remaining 900 µl of recovered cells by centrifugation at 5000 x g for 10 minutes. Remove most of the supernatant and resuspend pellet in remaining volume. Plate this volume onto the selective media.

Repeat the ligation with increased amounts of vector and/or insert and retransform.

Repeat vector dephosphorylation under conditions that will prevent "nibbling" the DNA termini.

Re-purify the vector and insert using the GENECLEAN® Turbo Kit (# 1102-200) or via standard phenol:chloroform extraction and precipitation to remove factors that could potentially interfere with ligation.

Transformation was inefficient or compromised

Review issues related to supercoiled plasmid transformation listed in Section 9.1.

9.3 I have too many colonies, or a bacterial lawn, on my transformation plate.

The efficiency of the protocol may have been underestimated

Repeat the procedure with less DNA if a lawn is observed.

Dilute the transformed cells or plate a smaller volume. Adjust the formula for calculating efficiency accordingly.

Determine transformation efficiency from the positive control to confirm kit operation and create a guideline for the expected number of transformants.

Some of the colonies may be bacterial contamination

Restreak the original bacterial stock on selective and nonselective media to confirm purity.

Prepare fresh competent cells using a pure bacterial culture.

Resterilize glassware used in the growth and cell preparation.

Effectiveness of the positive selection may be reduced

Analyze the growth media plate containing the positive control bacteria. It should not have any colonies. If it has colonies, check for contamination (see above) and repeat the transformation with fresh selective medium plates.

Ensure that the correct concentration of antibiotic was used to prepare the plates and that the plates were used shortly after preparation.

Ensure the agar was cooled to approximately 55°C (warm to the touch) prior to adding antibiotic and pouring plates.

Vector-contaminating background is higher than expected

Analyze the transformation plate representing the control vector without ligase. Colonies present on this plate indicate vector-only background that will contribute to background colonies in the vector with insert ligation transformation.

9.4 What if the colonies seem too large?

Larger than expected colonies are not a cause for concern. We recommend analyzing the colonies if all controls fall within expected results. Large colonies may be due to incubation at temperatures slightly higher than the recommended 37°C and/or incubation of the plates for greater than 18 - 24 hours. Variability in the bacterial plates (e.g. more media, less antibiotic or reduced antibiotic activity) may also account for larger-than-normal colony size.

9.5 What if there are two or more types of colonies?

Two types of colonies typically indicate contamination. This is especially true if they have different morphology, color or size (see Section 9.4). While it may be possible to pick a single colony that looks most like the anticipated bacteria and streak for isolation, it is best to repeat the transformation and ensure sterility.

It is important to note, however, that the presence of microcolonies that otherwise look like the larger colonies may indicate a phenotype associated with the transformed plasmid. Individual cells might respond differently to varying expression levels of a recombinant gene.

10. Recommended Reference Format for Publications

[Bacteria cell name] was transformed using the Transform & Grow™ Bacterial Transformation Kit (Qbiogene, Inc., CA).

11. References

1. Mandel, M. and A. Higa. (1970). *J. M. B.* 53:159 – 162.
2. Cohen, S. N. Chang, A. C. Y. and L. Hsu. (1972). *Proc. Natl. Acad. Sci., U. S. A.* 69:2110 – 2114.
3. Hanahan, D. (1983). *J. M. B.* 166:557 – 580. *Studies on transformation of Escherichia coli with plasmids.*
4. Inoue, H., Nojima, H., and H. Okayama. *Gene* (1990). *High efficiency transformation of Escherichia coli with plasmids.*
5. Chung, C. T., Niemela, S. L., and R. H. Miller. (1989). *Proc. Natl. Acad. Sci., U. S. A.* 86:2172 – 2175. *One-step preparation of competent Escherichia coli: Transformation and storage of bacterial cells in the same solution.*

12. Related Products

12.1 Plasmid Purification Kits

<u>Cat #</u>	<u>Description</u>	<u>Size</u>
2066-200	RapidPURE™ Plasmid Mini Kit	60 preps
2066-400	RapidPURE™ Plasmid Mini Kit	120 preps
2066-600	RapidPURE™ Plasmid Mini Kit	300 preps
2067-200	RapidPURE™ Plasmid Mini 96 Kit	96 preps
2067-400	RapidPURE™ Plasmid Mini 96 Kit	4 x 96 preps
2067-600	RapidPURE™ Plasmid Mini 96 Kit	10 x 96 preps
2069-400	Yeast RPM® Kit	100 preps
2000-200	MiniPrep Express™ Matrix	1,250 preps
2002-200	96well Prep Express	384 preps
2002-400	96well Prep Express	960 preps
2005-200	RapidPURE™ Plasmid Midi Kit	25 preps
2005-400	RapidPURE™ Plasmid Midi Kit	75 preps
2005-600	RapidPURE™ Plasmid Midi Kit	150 preps
2076-200	RapidPURE™ Plasmid Maxi GF Kit	20 preps
2074-200	RapidPURE™ Plasmid Maxi GF Endo Free Kit	10 preps
2078-200	RapidPURE™ Plasmid Giga Kit	12 preps

12.2 Other Transformation Kits and Tools

<u>Cat #</u>	<u>Description</u>	<u>Size</u>
2100-200	EZ-Yeast Transformation Kit	200 preps
2200-200	Alkali-Cation™ Yeast Transformation Kit	250 preps
2210-200	Yeast Spheroplast Transformation Kit	25 preps
3103-014	10x RbCl/CaCl Transformation Salts	150 ml
5000-552	Roll & Grow™ Plating Beads	2 Tubes

12.3 Molecular Biology Certified™ Growth Media

<u>Cat #</u>	<u>Description</u>	<u>Size</u>
3000-112	CIRCLEGROW® (Powder)	227 g (0.5 lb)
3000-165	CIRCLEGROW® (Pack)	10 x 0.5 L Pouches
3000-121	CIRCLEGROW® (Capsules)	250 Caps
3000-104	CIRCLEGROW® (Sterile Solution)	500 ml
3002-012	LB-Medium (Powder)	227 g (0.5 lb)
3002-065	LB-Medium (Pack)	10 x 0.5 L Pouches
3002-011	LB-Medium (Capsules)	227 g (0.5 lb)
3002-014	LB-Medium (Sterile Solution)	500 ml
3002-212	LB Agar Medium (Powder)	227 g (0.5 lb)
3002-265	LB Agar Medium (Pack)	10 x 0.5 L Pouches
3002-211	LB Agar Medium (Capsules)	227 g (0.5 lb)
3002-204	LB Agar Medium (Sterile Solid Solution)	500 ml
3103-011	SUPER-COMP Media (Capsules)	100 Capsules
3031-012	SOC Medium (Powder)	227 g (0.5 lb)

BIO 101® Systems is well known for providing an excellent selection of high-quality growth media in a variety of formulations. We specialize in formulations for yeast and bacterial genetics, and offer more than a thousand recipes and variations. Each product is subjected to extensive quantitative testing, and is Molecular Biology Certified™ through qualitative, molecular biology based tests such as cell density and plasmid yield. Choose from a wide variety of convenient packaging formats: Pre-mixed powders are available in capsule form, single-use pouches, or in bulk sizes. Liquid media, pre-poured plates, and custom packaging options are also available. If you prefer to make your own media, BIO 101® Systems can supply you with raw materials such as tryptones and peptones, yeast extract, a wide variety of sugars and salts, antibiotics, and other media additives.

13. Product Use Limitation & Warranty

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