molecular biology

Molecular cloning workflow solutions

Clone | Transform | Purify | Analyze



Clone

Transform

Purify

Analyze

Advance your cloning workflow

The molecular cloning workflow requires many steps to obtain the perfect clone for downstream applications. Advance your cloning workflow at every step—clone, transform, purify, and analyze to successfully produce a recombinant clone quickly.

This handbook is intended to guide you by providing technical information and clear choices across the cloning workflow with solutions designed for speed and simplicity.

With all of our cloning solutions, you can find what fits and get the results you need to move closer to the next step in your discovery.

Choose your simple, but powerful, cloning solutions and all necessary resources at **thermofisher.com/pcrandcloning**.

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Molecular cloning relies on recombinant DNA technologies to insert a DNA sequence of interest into a vector to generate a large number of copies of the recombinant vector.

Cloning has traditionally required restriction enzymes and a DNA ligase to form a new recombinant vector. However, recent cloning advancements, such as Invitrogen[™] TOPO[™] cloning, ligation-independent cloning, and gene synthesis, provide more efficient workflow solutions.

Cloning solutions overview

Cloning

For more than 25 years, we have provided superior tools for DNA cloning, continually improving upon traditional technologies and developing new ones. From restriction enzymes to complete cloning kits, we offer a comprehensive portfolio of tools and reagents that will help you save resources while obtaining high-quality cloned DNA to accelerate your next discovery.



Find more cloning tips and troubleshooting advice at thermofisher.com/cloningeducation and thermofisher.com/cloningsupport

Cloning solutions

TOPO cloning

Invitrogen[™] TOPO[™] PCR cloning technology was developed to help improve cloning efficiency, simplify protocol setup, and accommodate a wide range of PCR insert sizes. TOPO technology enables inserts with compatible ends to be readily joined to the vector in 5 minutes, without the need for additional ligation steps.

- **High-fidelity, blunt-end TOPO cloning**—designed for cloning PCR products amplified with high-fidelity polymerases; includes the *ccdB* gene for positive selection
- **TOPO TA cloning**—for fast, effective, direct cloning of *Taq* polymerase– amplified PCR products with overhangs or sticky ends
- **TOPO long-fragment cloning**—for easy, highly efficient cloning of extra-long PCR products using the linearized, topoisomerase I-activated pCR[™]-XL-2-TOPO vector



The three steps of TOPO PCR cloning technology.

Find out more at thermofisher.com/topocloning

TOPO cloning selection guide

Quickly find your TOPO cloning kit with our new interactive selection tool. Search by application, vector, or desired competent cells at **thermofisher.com/topoguide**

Gateway cloning system

To shuttle genes of interest between vectors, the Gateway cloning system offers site-specific, recombination-based cloning. With this easy-to-use choice for cloning in multiple expression systems, the insert's proper orientation and reading frame are maintained during shuttling using the Gateway vectors.

Find out more at thermofisher.com/gateway



The Gateway cloning reactions. This scheme shows the four types of plasmids and enzyme mixes involved in Gateway cloning reactions. Red arrows represent the fragment of interest. Adapted from Katzen F (2007) *Expert Opin Drug Discov* 2(4):571–589.

Found naturally in bacteria, restriction enzymes recognize and cleave specific DNA sequences, resulting in sticky ends (5' or 3' protruding ends) or blunt ends, which enable DNA inserts to be cloned into vectors with compatible ends. Star activity, buffer compatibility, and incomplete digestion are some common hurdles in restriction digestion.

FastDigest restriction enzymes

• Double and multiple digestions

in a universal buffer for any

combination of enzymes

To simplify cloning, we offer FastDigest enzymes, an advanced line of restriction enzymes that share buffer compatibility with downstream modifying enzymes. Benefits include:

• Complete digestion in 5–15 minutes

- No sequential digestions and buffer changes
- 176 unique specificities
 - Direct loading of the reaction mixture on gels

Plasmid DNA digested with NEB enzymes

9: Bgll

Doublestranded DA T C T T A A G C G Restriction enzyme A A T T C G C Restriction enzyme G C G

Find out more at thermofisher.com/fastdigest

Α

4: FastDigest Ndel

5: FastDigest Sall

Plasmid DNA digested with FastDigest enzymes



В

Figure 1. Comparison of digestion efficiencies of restriction enzymes. (A) FastDigest restriction enzymes digest plasmid DNA much more efficiently compared to the **(B)** NEB enzymes. In this experiment, using the NEB protocol, 1 µg of plasmid DNA was digested for ~15 minutes.

4: Ndel

5: Sall-HF

9: FastDigest BgIII

Type IIs restriction enzymes

A specific group of restriction enzymes, called type IIs endonucleases, cleave DNA outside of their recognition sequences. In combination with DNA ligase, type IIs restriction enzymes are utilized to drive the insertion of one or several DNA fragments into a recipient vector without the inclusion of residual restriction enzyme sites and other unwanted DNA sequences at fragment junctions (scarless cloning).

Find FastDigest type IIs enzymes at thermofisher.com/fastdigesttypeiis

For GeneArt Type IIs assembly kits go to thermofisher.com/typeiis

Cloning solutions (cont.)

GeneArt Gibson Assembly cloning kits

GeneArt Gibson Assembly cloning kits allow for simultaneous assembly of up to 15 large DNA fragments to precisely create very large constructs with no additional sequences in highly efficient reactions. This cloning method circumvents the need for multiple rounds of restriction enzyme analysis and digestion, DNA end repair, dephosphorylation, ligation, enzyme inactivation, and cleanup, and is a powerful tool in synthetic biology.

Benefits of GeneArt Gibson Assembly kits:

- Assembly of up to 15 fragments to build seamless clones
- Cloning efficiencies up to >95%
- Choice of complete kits with competent cells or master mixes

Find out more at thermofisher.com/gibsonassembly

Gibson Assembly HiFi kits:

Invitrogen[™] GeneArt[™] Gibson Assembly[®] HiFi kits offer a very cost-effective and efficient way of assembling smaller numbers of fragments.



Figure 1. GeneArt Gibson Assembly HiFi kits provide high cloning efficiency using single- to multiple-insert designs. Assembly of 1, 2, or 3 fragments of 1 kb in Invitrogen[™] pcDNA[™] 3.4 vector using Invitrogen[™] TOP10 competent cells. GeneArt Gibson Assembly HiFi kits are the most cost-effective and time-saving method for building large assemblies, particularly when used with GeneArt Strings DNA Fragments or 100% sequence-verified Invitrogen[™] GeneArt[™] Gene Synthesis.



Figure 2. Overview of the Gibson Assembly reaction, which relies on homologous recombination to assemble adjacent DNA fragments sharing end-terminal homology.

Gibson Assembly EX master mix and kit:

Invitrogen[™] GeneArt[™] Gibson Assembly[®] EX Master Mix and Invitrogen[™] GeneArt[™] Gibson Assembly[®] EX Cloning Kit provide high transformation efficiency for larger assemblies.



Figure 3. GeneArt Gibson Assembly EX kits provide high transformation efficiency options when using larger numbers of inserts. Assembly of 6, 8, and 10 fragments of 0.5 kb in pcDNA 3.4 vector transformed into Invitrogen[™] One Shot[™] TOP10 competent cells. GeneArt Gibson Assembly HiFi and EX technologies are available as master mixes or complete kits with chemically competent or electrocompetent cells, so you can have the flexibility of using your –20°C or –80°C freezer.



GeneArt Gene Synthesis

A reliable and cost-effective method for obtaining customized DNA constructs with 100% sequence accuracy, GeneArt Gene Synthesis offers:

- Synthetic, ready-to-transfect genes
- The ability to clone into many popular Invitrogen[™] vectors or your own custom vector
- Fully cloned, 100% sequence-verified genes ready for downstream applications
- Free optimization of a gene with Invitrogen[™] GeneOptimizer[™] software for maximum protein expression

Find out more at thermofisher.com/genesynthesis

GeneArt Strings DNA Fragments

A time-saving alternative to PCR, GeneArt Strings DNA Fragments are available in lengths up to 3 kb and are compatible with any downstream cloning method of choice, providing:

- Synthetic, ready-to-use DNA fragments
- DNA with your specified ends to facilitate the cloning method of choice
- No starting DNA required
- Free optimization of the gene with GeneArt GeneOptimizer software for maximum protein expression
- Option of Invitrogen[™] GeneArt[™] Strings[™] DNA Libraries with mixed, randomized nucleotides using full IUPAC codes

Find out more at thermofisher.com/strings



Transform

After the DNA fragment is cloned into a vector, competent bacteria are transformed with it to propagate sufficient quantities of the cloned DNA for downstream experiments. The choice of competent cells for transformation depends upon the transformation methods, strain genotypes, plasmid characteristics, and desired applications.

Find more transformation tips and troubleshooting advice at thermofisher.com/compcells and thermofisher.com/compcells-education



Competent cells

Choosing competent cells based on the application

We offer a broad portfolio of chemically competent and electrocompetent cells in a wide range of transformation efficiencies, formats, and strains. Use the decision tree below to select the best cells for your specific application area.



Competent cells (cont.)

Chemically competent cells

Chemically competent cells are treated with calcium chloride to facilitate attachment of the plasmid DNA to the cell membrane. The competent cells are heated in a water bath—this opens the pores of the cell membrane, allowing entry of the plasmid. Chemically competent cells are the best solution for general cloning and subcloning applications.

One Shot TOP10 cells

Chemically competent One Shot TOP10 *E. coli* cells are ideal for highefficiency cloning and plasmid propagation and are provided at a transformation efficiency of $1 \times 10^{\circ}$ cfu/µg plasmid DNA. They allow stable replication of high copy number plasmids and are the same competent cells that come with many of our cloning kits.

Electrocompetent cells

Electrocompetent cells are used in the electroporation process. Electrical pulses create pores that allow genetic material to permeate the bacterial membrane. The Invitrogen^T electrocompetent cell portfolio offers a variety of *E. coli* cells to reliably clone your DNA with high efficiency.

ElectroMAX DH10B cells

These electrocompetent *E. coli* cells offer the highest transformation efficiencies of >1 x 10¹⁰ cfu/µg plasmid DNA. Invitrogen[™] ElectroMAX[™] DH10B[™] cells are ideal for applications requiring high transformation efficiencies, such as with cDNA or gDNA library construction.



Chemical and electrocompetent transformation.

Find out more at thermofisher.com/compcells

Medium- and high-throughput transformation

Performing bacterial transformations one by one can be very time-consuming and create a bottleneck in your experimental workflow. There are times when medium- and high-throughput transformation options are desired. Invitrogen[™] MultiShot[™] chemically competent cells provide three flexible product formats to meet your throughput needs.

Find out more at thermofisher.com/multishot



StripWell format

- Medium-throughput option
- Twelve 8-tube strips
- Suitable for 1–96 transformations
- Five E. coli strains available

FlexPlate format

- High-throughput option
- 96-well plate separates into twelve 8-well segments
- Manual and automated platform transformations
- Six E. coli strains available





96-well plate

- Highest-throughput option
- Five 96-well plates
- Available with the One Shot TOP10 strain
- Stable replication of high copy number plasmids



Did you know?

Invitrogen[™] competent cells can be provided in custom configurations per your request. Large and custom volumes, as well as multiple formats, are at your fingertips. Simply email us at customorders@thermofisher.com

Transform

Purify

Once the bacterial cells are transformed with the vector, it is critical to recover plasmid DNA using plasmid purification products that are optimized to provide maximum yield, purity, and integrity.

Find more plasmid purification tips and troubleshooting resources at **thermofisher.com/plasmid** and **thermofisher.com/plasmidsupport**

Plasmid purification solutions

Purify

Plasmid purification is a basic technique necessary for generating recombinant clones. Thermo Scientific[™] GeneJET[™] and Invitrogen[™] PureLink[™] plasmid purification kits have been designed and developed with your experimental specifications, plasmid purity, and throughput demands in mind. Use the decision tree below to select the right plasmid purification kit based on the intended application.



Find out more at **thermofisher.com/plasmid**

Solutions for molecular applications

GeneJET and PureLink kits

Robust purification of plasmid DNA, in the amount and purity required for the downstream application of interest, is a key step of the cloning workflow. During the construction of plasmid DNA and subsequent verification (sequencing, enzymatic digestion), small-scale plasmid DNA preparations are generally used (see the table below). Silica membrane–based columns such as GeneJET and PureLink products are ideal due to their fast and simple purification workflows.

These kits offer the following benefits:

- High yields of low-throughput, molecular-grade plasmid DNA
- Ideal for basic molecular biology applications, including PCR, sequencing, cloning, and transcription
- Thermo Scientific[™] GeneJET[™] Plasmid Miniprep, Midiprep, and Maxiprep kits offer the best overall value and ease of use, and the fastest processing (for conventional pellet method)
- The Invitrogen[™] PureLink[™] Plasmid Miniprep Kit offers a slightly higher yield and comes more complete (elution tubes plus optional buffers included) in mini-scale column format

Find out more at thermofisher.com/genejet and thermofisher.com/purelink

Scale considerations for plasmid purification

Parameter	Miniprep	Midiprep	Maxiprep	Megaprep	Gigaprep
Bacterial culture volume	1–5 mL	10–50 mL	100–500 mL	500 mL-2.5 L	2.5–5 L
Plasmid yield	Up to 40 µg	Up to 300 µg	Up to 1 mg	Up to 4 mg	Up to 15 mg



Solutions for transfection applications



PureLink kits

For experiments involving cells and animal models, endotoxin levels are one of a scientist's primary concerns. Endotoxins can induce immune responses, resulting in suboptimal transfection and toxicity in many cell lines, and negatively influence protein expression in sensitive cells. To enable successful *in vitro* and *in vivo* studies, Invitrogen[™] PureLink[™] HiPure, Fast Low-Endotoxin, and Expi Endotoxin-Free kits are ideal for applications that require high purity of plasmids and low-to-"zero" levels of endotoxins.

Find out more at thermofisher.com/plasmid





Figure 5. Higher yields of low-endotoxin, advanced transfection-quality plasmid DNA obtained with a PureLink Fast Low-Endotoxin purification kit than with a comparable kit from another supplier. Two high-copy plasmids with different backbones were purified using the PureLink Fast Low-Endotoxin and Supplier Q maxiprep kits as described in the product manuals. Plasmid yields are shown. Endotoxin values (EU/µg) were measured using an Endosafe[™]-PTS instrument (Charles River Laboratories) and are provided only for the PureLink Fast Low-Endotoxin preparations.

Figure 6. Higher yields of endotoxin-free, advanced transfection-quality plasmid DNA obtained with a PureLink Expi purification kit than with a comparable kit from another supplier. Two high-copy plasmids with different backbones were purified using the PureLink Expi Endotoxin-Free and Supplier Q maxiprep kits as described in the product manuals. Plasmid yields are shown. Endotoxin values (EU/µg) were measured using the EndoSafe-PTS test and are provided only for the PureLink Expi preparations.

2

<0.05 EU/µg

PureLink Expi kit

Supplier Q EndoFree kit

Maxi

Plasmid

<0.05 EU/ua

1

1.5

1.2

0.9

0.6

0.3

0



Choosing the right tools for your cloning analysis can significantly improve and accelerate results, enabling you to address downstream applications sooner. Explore our precast analysis options, do-it-yourself reagents, and isolation kits to find the right workflow solution for your cloning analysis needs.

Find more nucleic acid analysis tips and troubleshooting resources at thermofisher.com/na-electrophoresis-education



Sample analysis solutions

Identifying the appropriate gel type and gel concentration for your analysis is an essential step that will streamline the separation of nucleic acids. Find out more about our convenient reagents for sample analysis using agarose gel electrophoresis, including pour-your-own Invitrogen[™] UltraPure[™] agarose reagents and hassle-free, precast Invitrogen[™] E-Gel[™] agarose gels in this section.



Find out more at **thermofisher.com/egel**

Sample analysis solutions (cont.)

E-Gel precast gels

Using precast agarose gels can simplify the nucleic acid electrophoresis workflow. E-Gel precast gels are self-contained and ready for use, with the agarose and the DNA stain packaged in a disposable cassette. The use of in-gel Invitrogen[™] SYBR[™] Safe DNA Gel Stain and a blue-light transilluminator minimizes DNA damage and improves cloning efficiency compared to conventional methods. There are no gels to pour, buffers to make, staining or destaining steps to perform, or gel boxes to assemble. Just load your samples and start the run.



Sample analysis in three simple steps-load, run, and analyze.

Find out more at thermofisher.com/egel

E-Gel Power Snap Electrophoresis System

To help reduce workflow time, both the Invitrogen[™] E-Gel[™] Power Snap Electrophoresis System (11- and 22-wells) and the Invitrogen[™] E-Gel[™] Power Snap Plus Electrophoresis System (11-, 22-, 48- and 96-wells) integrate rapid, real-time acid analysis with high-resolution image capture.

Find out more at **thermofisher.com/powersnap** 20

Electrophoresis reagents

For pour-your-own agarose gels, choosing high-quality agarose, optimized DNA ladders, and improved DNA stains can help you achieve optimal electrophoresis results.



UltraPure reagents for electrophoresis

Invitrogen[™] UltraPure[™] reagents are specifically formulated to meet your nucleic acid analysis and purification needs. UltraPure agarose and reagents are made from highly pure biochemicals for maximum reliability and superior performance.

Find out more at thermofisher.com/ultrapure

DNA stains

Detection of nucleic acid samples in gels can be improved using fluorescent dyes that are safer and more sensitive than ethidium bromide. SYBR Safe and Invitrogen[™] SYBR[™] Gold Nucleic Acid Gel stains provide greater safety and sensitivity with lower background fluorescence than the conventional ethidium bromide stain.

Find out more at thermofisher.com/stains

DNA ladders

Invitrogen[™] DNA Ladders are available for a wide variety of size ranges, applications, and formats. SYBR Safe stain has lower background fluorescence than the conventional ethidium bromide stain. They accurately estimate size and mass of double-stranded, single-stranded, or supercoiled DNA fragments.

Find out more at thermofisher.com/ladders



Sample isolation solutions

Whether isolating a specific DNA fragment from complex PCR mixtures or recovering bands from agarose gels, we have solutions that will meet your needs. Isolate DNA fragments that are ready for a variety of downstream applications like sequencing, PCR, transcription, cloning, and labeling.



DNA load = 25 ng/lane Lane 1: 1 kb Plus DNA Ladder Lane 2: 100 bp amplicon, unpurified Lane 3: 100 bp amplicon, purified Lane 4: 400 bp amplicon, purified Lane 6: 965 bp amplicon, unpurified Lane 7: 965 bp amplicon, unpurified Lane 8: 2 kb amplicon, unpurified Lane 9: 2 kb amplicon, purified Lane 10: 5.4 kb amplicon, unpurified

$\label{eq:amplification} \mbox{ Amplification of DNA isolated using the Invitrogen^{\tiny $\ensuremath{\mathbb{T}}$}\mbox{ PureLink}^{\tiny $\ensuremath{\mathbb{T}}$}\mbox{ Quick Gel Extraction Kit.}$

PCR amplicons varying in size from 100 bp to 5.4 kb were prepared using recombinant Invitrogen[™] *Taq* DNA Polymerase. A portion of each PCR reaction was run on a 1% Invitrogen[™] UltraPure[™] Agarose gel (data not shown), and amplicon bands were excised and extracted using the PureLink Quick Gel Extraction Kit. The Invitrogen[™] 1 Kb Plus DNA Ladder was used in this gel.



Kits for DNA cleanup

Product	Protocol time (min)	DNA cleanup application	Format	Elution volume (μL)	Quantity
GeneJET PCR Purification Kit	5	PCR cleanup	Silica spin or vacuum column	50	50 preps 250 preps
PureLink Quick Gel Extraction Kit	<30	Gel extraction	Silica spin or vacuum column	30–100	50 preps 250 preps

Resources

A comprehensive portfolio of educational resources, frequently asked questions, and mobile apps are now available to help you reach new heights in your research.

Find more cloning tips and troubleshooting advice at thermofisher.com/cloningeducation and thermofisher.com/cloningsupport

Frequently asked questions

Below are some common questions and answers to help you start or troubleshoot your molecular biology experiments.

Clone What are some of the prerequisites for TOPO cloning? Please consider the following before you do TOPO cloning:

- TOPO cloning cannot ligate DNA with a 5' phosphate group.
- TOPO cloning will decrease in efficiency inversely with the size of the insert (>3 kb) unless you use the TOPO XL cloning kit.
- TOPO vectors contain different antibiotic resistance markers, which should be considered before purchase.

Are GeneArt Gibson Assembly kits available with electrocompetent cells?

Yes, both the GeneArt Gibson Assembly HiFi and EX configurations are available with electrocompetent cells. Go to **thermofisher.com/gibsonassembly** to see the full list of available products.

What is the main difference between GeneArt Strings DNA Fragments and GeneArt Gene Synthesis?

GeneArt Strings DNA Fragments are custom-made, uncloned, double-stranded linear DNA fragments. GeneArt Gene Synthesis is a service offered for chemical synthesis, cloning, and sequence verification of genetic sequences.

Transform

Can you explain the significant differences between the TOP10, DH5a, and Mach1 strains that you have for the cloning reactions? DH5a cells are commonly used for routine cloning, but are *mcr/mrr*⁺ and therefore not recommended for genomic cloning. The TOP10 competent cells, on the other hand, are *mcr/mrr*⁻, and therefore are a good choice for routine cloning and can be used for cloning of methylated DNA, such as eukaryotic genomic DNA. Our Mach1 strain is the fastest-growing cloning strain that is T1 phage-resistant.

Purify

What growth conditions do you recommend for *E. coli* for isolating plasmid DNA using your plasmid isolation kits?

We typically recommend growing *E. coli* up to $A_{600} = 1-1.5$ (~1 x 10° cells/mL) in LB broth.

Can I elute my plasmid from PureLink columns with water instead of TE?

For any silica columns, elution with water is generally possible. However, a buffer is preferred for stability and accuracy of absorbance readings, as pure water can have a very low pH (4–5).

Analyze

I want to pour my own gels. Which agarose should I use?

The UltraPure Agarose is standard, melting-point agarose designed for routine separation and analysis of DNA and RNA fragments in the 500–23,000 nt range. Invitrogen[™] UltraPure[™] Agarose-1000 is a specialized agarose that provides higher resolution of PCR fragments and other short DNA fragments. We also offer an Invitrogen[™] UltraPure[™] Low Melting Point Agarose, which is ideal for resolving and isolating DNA fragments from 10 to 1,000 bp with a low melting temperature of 65°C or less.

Mobile apps



DailyCalcs-science calculator

The DailyCalcs app turns your phone into a science calculator to help simplify everyday tasks in the lab. The app features eight calculators: molarity, dilution, formula weight, transfection, unit conversions, culture vessel data, media conversions, and specific productivity.



Instrument Connect—remote monitoring Instrument Connect allows you to view instrument status, monitor or schedule a run, and more on any cloud-enabled instrument, including the Applied Biosystems[™] ProFlex[™], SimpliAmp[™], and MiniAmp[™] PCR instruments.



PCR Quest—match-3 lab game Test your PCR knowledge with our lab game— PCR Quest—where you travel from lab to lab crushing the world's toughest diseases. Download at thermofisher.com/pcrquest

Custom and OEM solutions

As a leading supplier of molecular biology reagents, plastics, and instruments, we offer customizable manufacturing solutions used by companies in developing next-generation molecular assays. Regardless of where you are in your assay development, we have off-the-shelf or custom solutions to help you achieve your goals. Partner with an experienced supplier that understands both raw materials and new technologies—a market leader with a dedicated diagnostics partnering business that brings value beyond products.

What do our OEM solutions mean to you?

- Customization of products and services
- Consultation, partnership, and expertise
- Negotiated business terms
- Warranties and indemnification
- Commercial-use rights and obligations
- Risk and liability management



Find out more at thermofisher.com/oemmolecular

Find out more at thermofisher.com/mobileapps

Ordering information

Product	Quantity	Cat. No.
Clone		
FastDigest BamHI	800 reactions	FD0054
FastDigest Bcul	50 reactions	FD1254
FastDigest BshTl	20 reactions	FD1464
FastDigest DpnI	100 reactions	FD1704
FastDigest EcoRI	800 reactions	FD0274
FastDigest Kpnl	300 reactions	FD0524
FastDigest Notl	50 reactions	FD0594
FastDigest Sall	200 reactions	FD0644
FastDigest Xbal	300 reactions	FD0684
FastDigest Xhol	400 reactions	FD0694
FastDigest Esp3I (BsmBI) (type IIs)	20 reactions	FD0454
FastDigest Bpil (Bbsl) (type IIs)	20 reactions	FD1014
FastDigest Eco31I (Bsal) (type IIs)	100 reactions	FD0294
TOPO TA Cloning Kit for Subcloning, without competent cells	10 reactions	451641
Zero Blunt TOPO PCR Cloning Kit, without competent cells	10 reactions	451245
Zero Blunt TOPO PCR Cloning Kit for Sequencing, with One Shot TOP10 Chemically Competent E. coli	25 reactions	K287520
pENTR/D-TOPO Cloning Kit, with One Shot TOP10 Chemically Competent E. coli	20 reactions	K240020
TOPO XL-2 Complete PCR Cloning Kit, with One Shot OmniMAX 2 T1 [®] Chemically Competent E. coli	20 reactions	K8050-20
TOPO TA Cloning Kit for Subcloning, without competent cells	10 reactions	451641
Gateway LR Clonase II Enzyme Mix	20 reactions	11791020
MultiSite Gateway Pro Plus	20 reactions	12537100
MultiShot FlexPlate Stbl3 Chemically Competent E. coli	1 plate	C7381201
PCR Cloning System with Gateway Technology with pDONR 221 and OmniMAX 2 Competent Cells	20 reactions	12535029
GeneArt Gibson Assembly HiFi Cloning Kit, chemically competent cells	10 reactions	A46624
GeneArt Gibson Assembly HiFi Master Mix	10 reactions	A46627
GeneArt Gibson Assembly EX Cloning Kit, chemically competent cells	10 reactions	A46633
GeneArt Gibson Assembly EX Master Mix	10 reactions	A46635
GeneArt Gene Synthesis	thermofisher.com/gene	synthesis

Ordering information (cont.)

Product	Quantity	Cat. No.
Transform		
One Shot TOP10 Chemically Competent <i>E. coli</i>	20 reactions	C404003
MultiShot FlexPlate TOP10 Chemically Competent E. coli	1 plate	C4081201
MAX Efficiency DH5a Competent Cells	200 µL	18258012
MultiShot FlexPlate DH5α T1 [®] Chemically Competent <i>E. coli</i>	1 plate	C4481201
ElectroMAX DH10B Cells	100 µL	18290015
MAX Efficiency Stbl2 Competent Cells	5 x 200 µL	10268019
One Shot Stbl3 Chemically Competent E. coli	20 x 50 µL	C737303
MultiShot FlexPlate Stbl3 Chemically Competent E. coli	1 plate	C7381201
Purify		
	4 preps	A33073
PureLink Expi Endotoxin-Free Maxi Plasmid Purification Kit	10 preps	A31217
	25 preps	A31231
PureLink Expi Endotoxin-Free Mega Plasmid Purification Kit	4 preps	A31232
PureLink Expi Endotoxin-Free Giga Plasmid Purification Kit	2 preps	A31233
PureLink Expi Endotoxin-Free Buffer Set	1 set	A33074
Pural ink Fast Low-Endotoxin Midi Plasmid Purification Kit	25 preps	A35892
	50 preps	A36227
Purel ink Fast Low-Endotoxin Maxi Plasmid Purification Kit	25 preps	A35895
	50 preps	A36228
Purel ink HiPure Plasmid Mininren Kit	25 preps	K210002
	100 preps	K210003
Purel ink HiPure Plasmid Midioren Kit	25 preps	K210004
	100 preps	K210005
PureLink HiPure Plasmid Maxiprep Kit	10 preps	K210006
	25 preps	K210007
PureLink HiPure Expi Plasmid Megaprep Kit	4 preps	K210008XP
PureLink HiPure Expi Plasmid Gigaprep Kit	2 preps	K210009XP
PureLink Quick Plasmid Miniprep Kit (molecular grade)	50 preps	K210010
	250 preps	K210011
GeneJET Plasmid Midiprep Kit	25 preps	K0481
	100 preps	K0482
	10 preps	K0491
Geneje i Plasmia Maxiprep Kit	25 preps	KU492
Cana IET Diagnaid Miniayan Kit	50 preps	KU5U2
Geneje i Masmia Miniprep Kit	250 preps	KU5U3

Ordering information (cont.)

Product	Quantity	Cat. No.
Analyze		
UltraPure Ethidium Bromide	10 mL	15585011
UltraPure Agarose	100 g	16500100
UltraPure DNase/RNase-Free Distilled Water	10 x 500 mL	10977023
UltraPure TAE Buffer 10X	4 L	15558026
TrackIt 1 Kb Plus DNA Ladder	50 µg	10488085
SYBR Safe DNA Gel Stain	400 µL	S33102
E-Gel Agarose Gels EX, 1%	10 gels	G401001
E-Gel Agarose Gels EX, 2%	10 gels	G401002
E-Gel Agarose Gels EX, 4%	10 gels	G401004
E-Gel Agarose Gels with SYBR Safe DNA Gel Stain, 1%	10 gels	A42100
E-Gel Agarose Gels with SYBR Safe DNA Gel Stain, 2%	10 gels	A42135
E-Gel Agarose Gels with SYBR Safe DNA Gel Stain, 4%	10 gels	A42136
E-Gel EX Double Comb Agarose Gels, 1%	10 gels	A42345
E-Gel EX Double Comb Agarose Gels, 2%	10 gels	A42346
E-Gel Double Comb Agarose Gels with SYBR Safe DNA Gel Stain, 1%	10 gels	A42347
E-Gel Double Comb Agarose Gels with SYBR Safe DNA Gel Stain, 2%	10 gels	A42348
E-Gel CloneWell II Agarose Gels with SYBR Safe, 0.8%	10 gels	G661818
E-Gel SizeSelect II Agarose Gels, 2%	10 gels	G661012
E-Gel NGS 0.8% Agarose Gels	10 gels	A25798
E-Gel Go! Agarose Gels, 1%	10 gels	G441001
E-Gel Go! Agarose Gels, 2%	10 gels	G441002
E-Gel 48 Agarose Gels with SYBR Safe DNA Gel Stain, 1%	8 gels	G820801
E-Gel 48 Agarose Gels with SYBR Safe DNA Gel Stain, 2%	8 gels	G820802
E-Gel 48 Agarose Gels with SYBR Safe DNA Gel Stain, 4%	8 gels	G820804
E-Gel 96 Agarose Gels with SYBR Safe DNA Gel Stain, 1%	8 gels	G720801
E-Gel 96 Agarose Gels with SYBR Safe DNA Gel Stain, 2%	8 gels	G720802
PureLink Quick Gel Extraction Kit	50 preps	K210012
GeneJET PCR Purification Kit	50 preps	K0701

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