Reagents and Tools for Molecular Cloning

CLONE WITH CONFIDENCE®

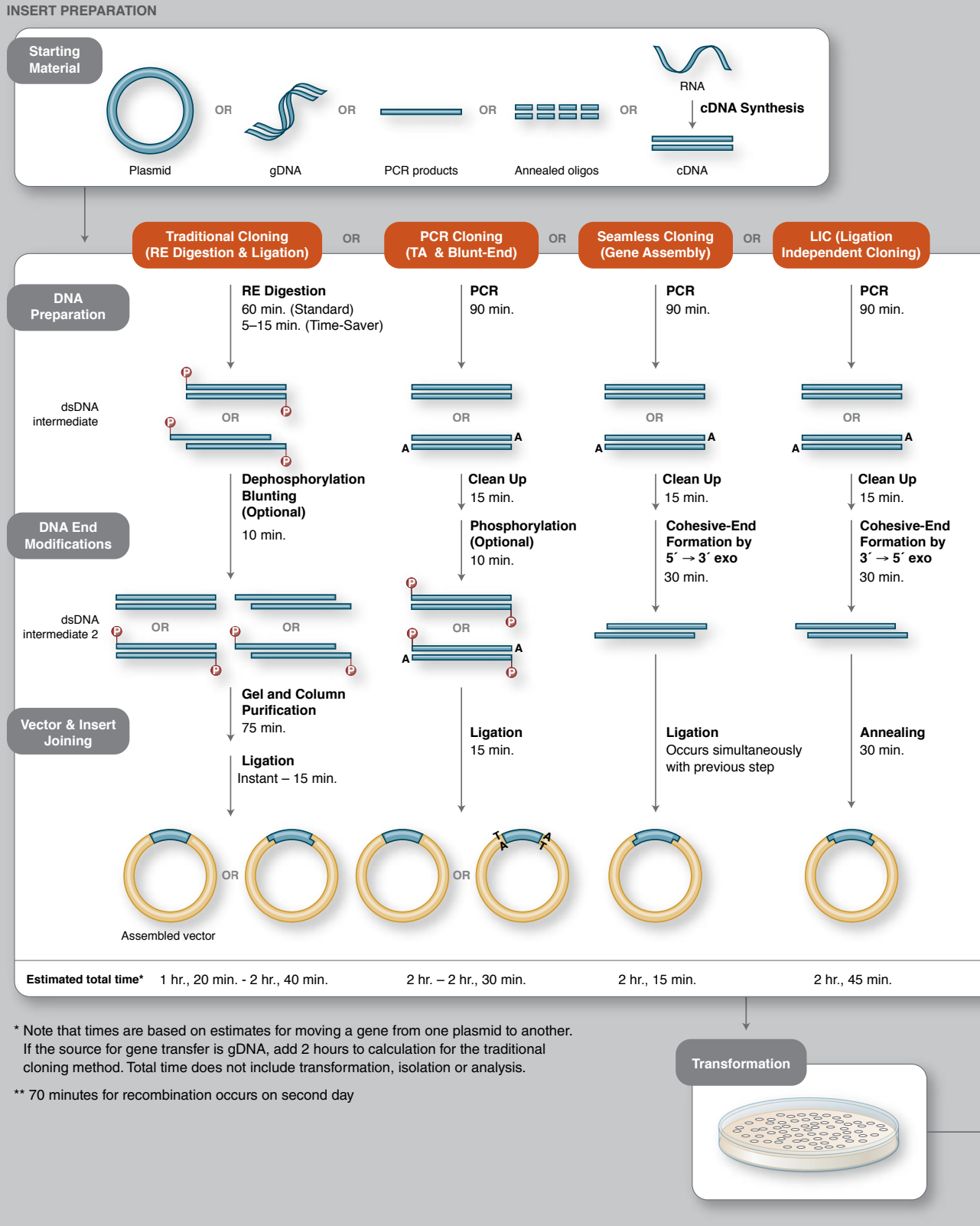
NEW ENGLAND BioLabs®

be INSPIRED
drive DISCOVERY
stay GENUINE

Clone with confidence®

Built on over 40 years of experience, New England Biolabs® provides you with the quality reagents and technical support that you need to take your cloning experiments to the next level. With reagents for each step in the cloning workflow, NEB® products deliver quality and performance, so that you can clone with confidence.

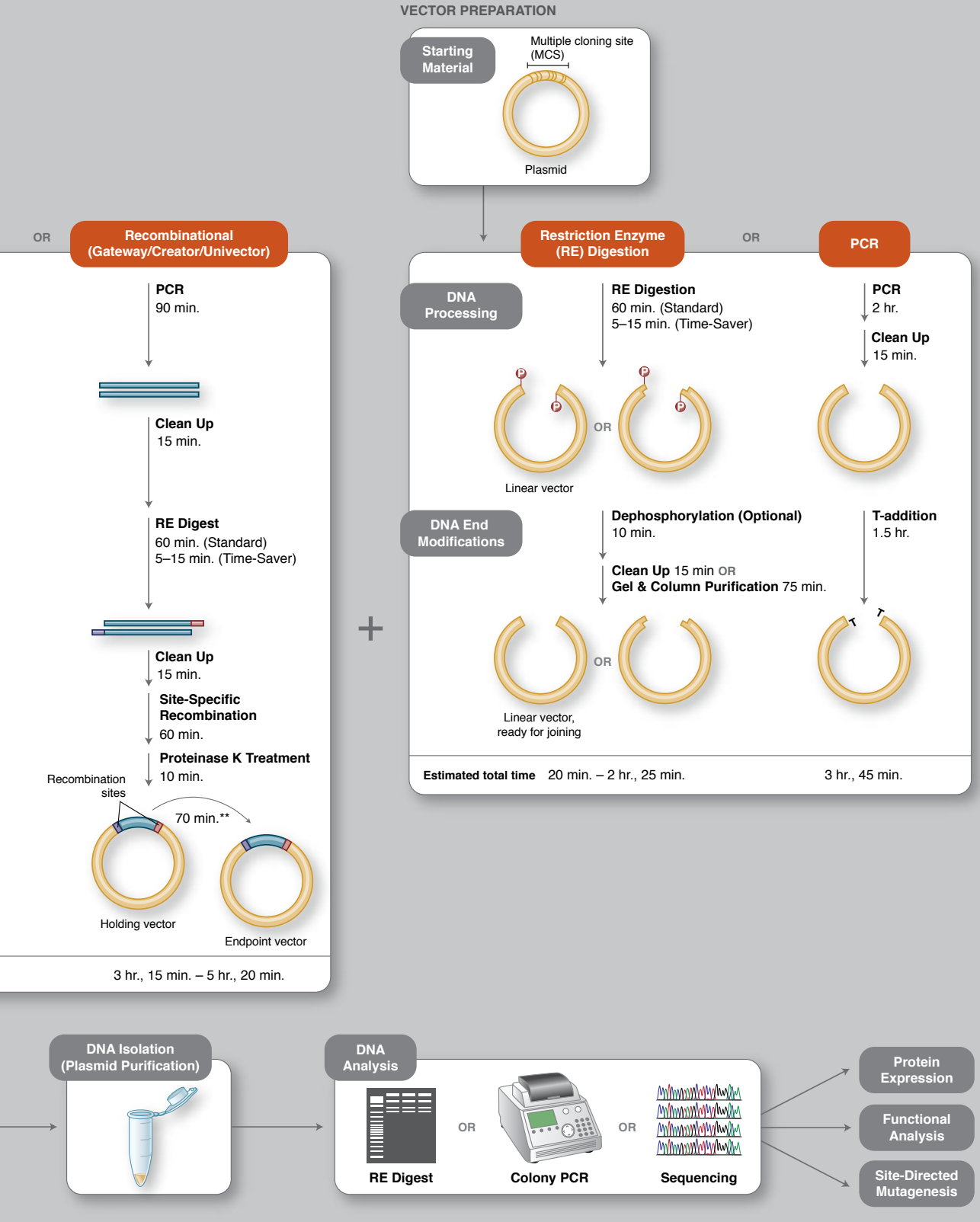
Cloning Workflow Comparison



* Note that times are based on estimates for moving a gene from one plasmid to another. If the source for gene transfer is gDNA, add 2 hours to calculation for the traditional cloning method. Total time does not include transformation, isolation or analysis.

** 70 minutes for recombination occurs on second day

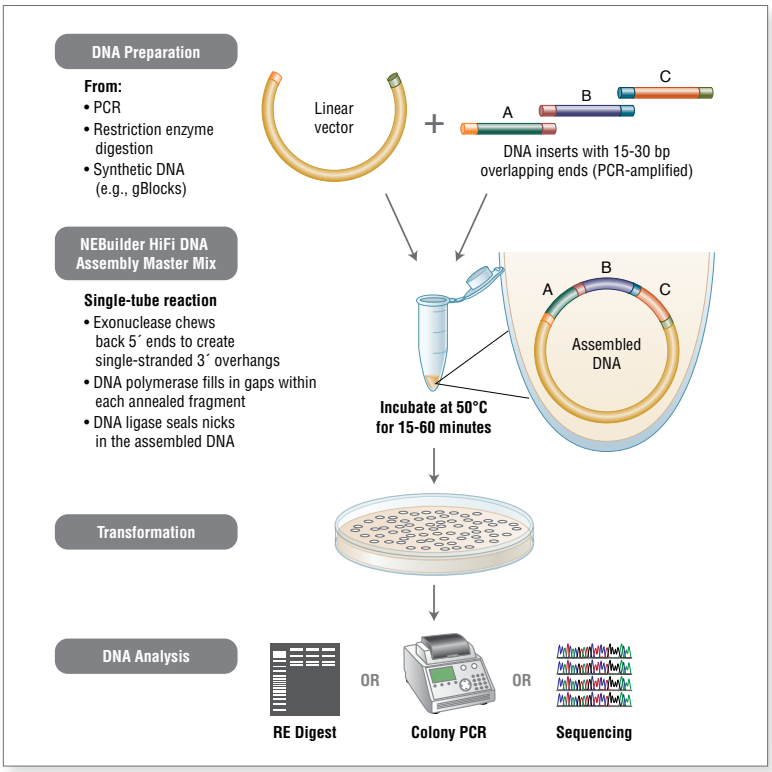
The figure below compares the various cloning methodologies. To help get you started with your experiment, we have included selection charts for each step in the workflows, as well as recommended products and tips to ensure successful cloning experiments. To learn more about the full portfolio of products available, visit **CloneWithNEB.com**.



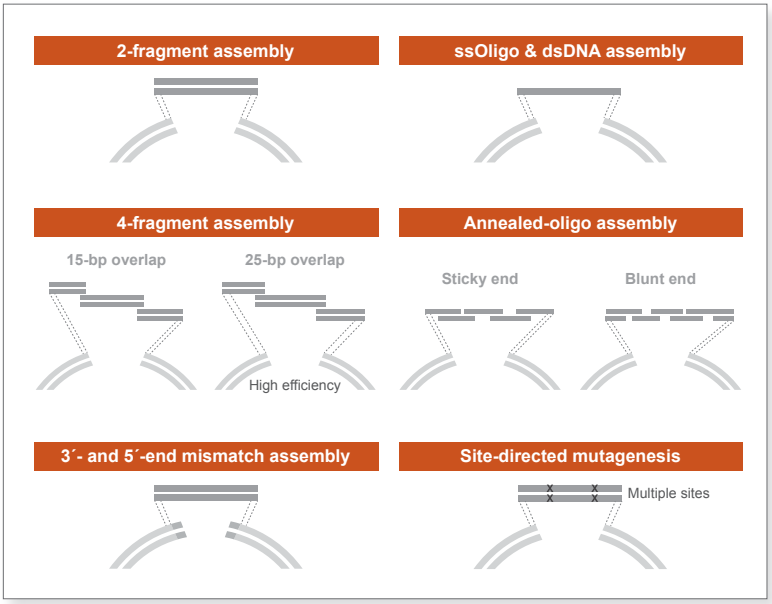
NEBuilder® HiFi DNA Assembly

NEBuilder HiFi DNA Assembly enables virtually error-free joining of DNA fragments, even those with 5´- and 3´-end mismatches. Available with and without competent *E. coli*, this flexible kit enables simple and fast seamless cloning utilizing a proprietary high-fidelity polymerase. Make NEBuilder HiFi your first choice for DNA assembly and cloning.

Overview of the NEBuilder HiFi DNA Assembly cloning method



NEBuilder HiFi DNA Assembly
can be used for a variety of DNA assembly methods.



RECOMMENDED PRODUCTS

NEBuilder HiFi DNA Assembly Cloning Kit (NEB #E5520)

NEBuilder HiFi DNA Assembly Master Mix (NEB #E2621)

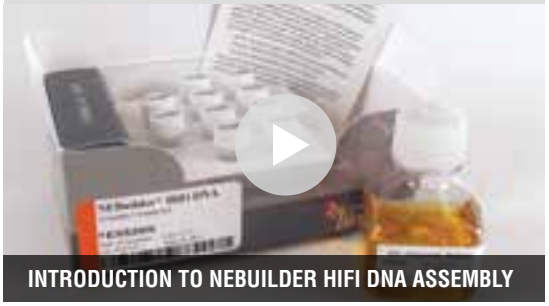
NEBuilder HiFi DNA Assembly Bundle for Large Fragments (NEB #E2623)

- Simple and fast seamless cloning
- Increased number of successful assembly products, particularly for longer or greater numbers of fragments
- Flexible sequence design, with no need to engineer cloning site
- No PCR cleanup step required
- Complex assembly achieved in an hour
- Less screening/re-sequencing of constructs, virtually error-free, high-fidelity assembly
- Use in successive rounds of assembly; removes 5´ and 3´ restriction enzyme mismatches
- Bridge two ds-fragments with a synthetic ss-DNA oligo
- Use one system for both "standard-size" cloning and larger gene assembly products, up to 6 fragments
- DNA can be used immediately for transformation or as template for PCR or RCA
- Adapts for multiple DNA manipulations, including site-directed mutagenesis
- No licensing fees from NEB for NEBuilder products

TOOLS & RESOURCES

Visit NEBuilderHiFi.com to find:

- Online tutorials to help with assembly and primer design
- Application notes utilizing NEBuilder HiFi
- Access to **NEBuilder Assembly Tool**, our online primer design tool and **NEBioCalculator®** for scientific calculations



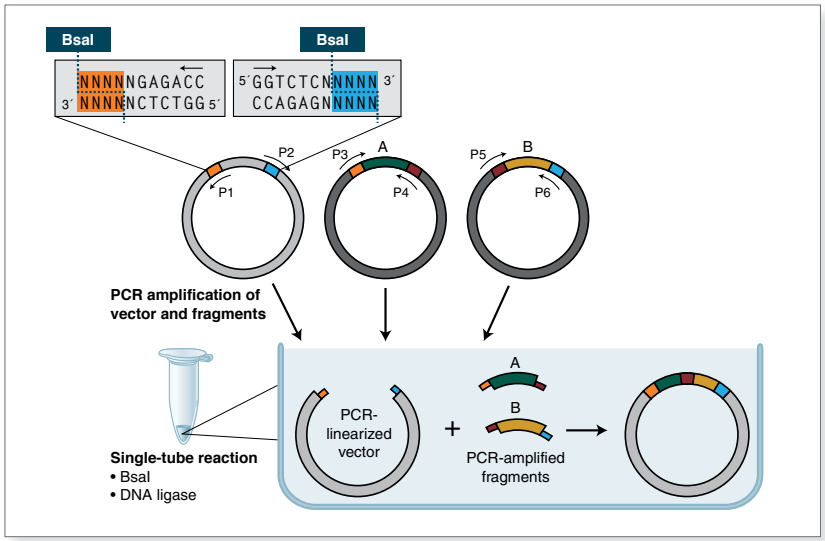
For help with designing primers, try NEBuilder Assembly Tool at **NEBuilder.neb.com**

NEB Golden Gate Assembly

The efficient and seamless assembly of DNA fragments, commonly referred to as Golden Gate assembly (1,2), has its origins in 1996 when, for the first time, it was shown that multiple inserts could be assembled into a vector backbone using only the sequential (3) or simultaneous (4) activities of a single Type IIS restriction enzyme and T4 DNA Ligase. This method can be accomplished using Type IIS restriction enzymes, such as BsaI, and can also be used for the cloning of single inserts into a vector. The method is efficient and can be completed in one tube in as little as 5 minutes for single inserts, or can utilize cycling steps for multiple inserts.

The NEB Golden Gate Assembly Mix incorporates digestion with BsaI and ligation with T4 DNA Ligase into a single reaction, and can be used to assemble up to 10 fragments in a single step.

NEB Golden Gate Assembly workflow



In its simplest form, Golden Gate Assembly requires a Type IIS recognition site, in this case, BsaI (GGTCTC), added to both ends of a dsDNA fragment. After digestion, these sites are left behind, with each fragment bearing the designed 4-base overhangs that direct the assembly.

RECOMMENDED PRODUCTS

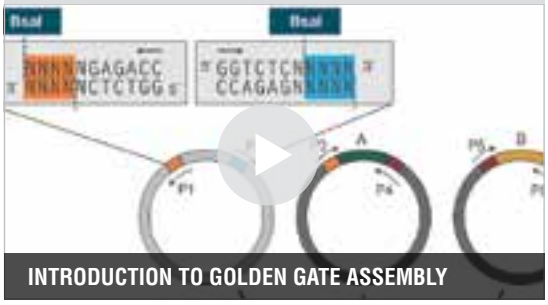
NEB Golden Gate Assembly Mix (NEB #E1600)

- Seamless cloning – no scar remains following assembly
- Ordered assembly of up to 10 fragments in a single reaction
- Efficient with regions with high GC content and areas of repeats
- Compatible with a broad range of fragment sizes (< 100 bp to > 15 kb)
- Includes destination vector for efficient BsaI-based Golden Gate assemblies

TOOLS & RESOURCES

Visit www.neb.com/GoldenGate to find:

- Publications and protocols related to Golden Gate Assembly
- Access to **NEB Golden Gate Assembly Tool**, our online assembly tool
- A list of Type IIS restriction enzymes that can be used with **NEB Golden Gate Assembly**



Speed up your experimental design with our online assembly tool at **NEBGoldenGate.neb.com**

NEB Gibson Assembly®

Gibson Assembly enables multiple, overlapping DNA fragments to be joined in a single-tube isothermal reaction, with no additional sequence added (scar-less).

RECOMMENDED PRODUCTS

Gibson Assembly Cloning Kit (NEB #E5510)

Gibson Assembly Master Mix (NEB #E2611)

Visit NEBGibson.com for more information.

SGHDNA

Some components of this product are manufactured by New England Biolabs, Inc. under license from Synthetic Genomics, Inc.

Comparison of DNA Assembly Reaction Types

	NEBuilder HiFi DNA Assembly		NEB Gibson Assembly		In-Fusion® HD	
Assembly reaction types	**Assembly efficiency**	**Covalently sealed?***	**Assembly efficiency**	**Covalently sealed?***	**Assembly efficiency**	**Covalently sealed?***
2-fragment assembly						
No mismatch	+++	Yes	++	Yes	++	No
3′- and 5′-end mismatch	+++	Yes	++	Yes	X	No
4-fragment assembly						
15-bp overlap & no mismatch	+++	Yes	++	Yes	++	No
25-bp overlap & no mismatch	+++	Yes	++	Yes	++	No
Oligo assembly						
3′- and 5′-overhang	+++	Yes	++	Yes	X	No
Blunt end & no mismatch	+++	Yes	++	Yes	X	No
ssOligo & vector	+++	Yes	NP	Yes	X	No

* Assembled products are treated with T5 exonuclease followed by PCR. Only covalently sealed products resistant to T5 exonuclease digestion can serve as templates for PCR and yield PCR product.

+++ Performs best; recommended
++ Performs well; but other product(s) perform better
\+ Performs, but not recommended
X Does not perform
NP Experiment not performed

DNA Assembly Selection Chart

	NEBuilder HiFi DNA Assembly	NEB Gibson Assembly	NEB Golden Gate Assembly Mix	USER™ Enzyme
PROPERTIES				
Removes 5′ or 3′ End Mismatches	★★★	★	N/A	N/A
Assembles with High Fidelity at Junctions	★★★	★★	★★★	★★★
Tolerates Repetitive Sequences at Ends	★	★	★★★	★★★
Generates Fully Ligated Product	★★★	★★★	★★★	NR
Joins dsDNA with Single-stranded Oligo	★★★	★★	NR	NR
Assembles with High Efficiency with Low Amounts of DNA	★★★	★★	★★	★★
Accommodates Flexible Overlap Lengths	★★★	★★★	★	★★
APPLICATIONS				
Simple Cloning (1-2 Fragments)	★★★	★★★	★★★	★★★
4-6 Fragment Assembly	★★★	★★★	★★★	★★★
>6 Fragment Assembly	★★★	★★	★★★	★★★
Template Construction for In vitro Transcription	★★★	★★★	★★★	★
Synthetic Whole Genome Assembly	★★★	★	★	★
Multiple Site-directed Mutagenesis	★★★	★★	★★	★★
Library Generation	★★	★★	★★	★★
Pathway Engineering	★★★	★★	★★	★★★
TALENs	★★	★★	★★★	★★
Short Hairpin RNA Cloning (shRNA)	★★★	★★	★	★
gRNA Library Generation	★★★	★★	★	★
Large Fragment (>10 kb) Assembly	★★★	★★★	★★★	★★
Small Fragment (<100 bp) Assembly	★★★	★	★★★	★★★
Use in Successive Rounds of Restriction Enzyme Assembly	★★★	★	NR	★

KEY

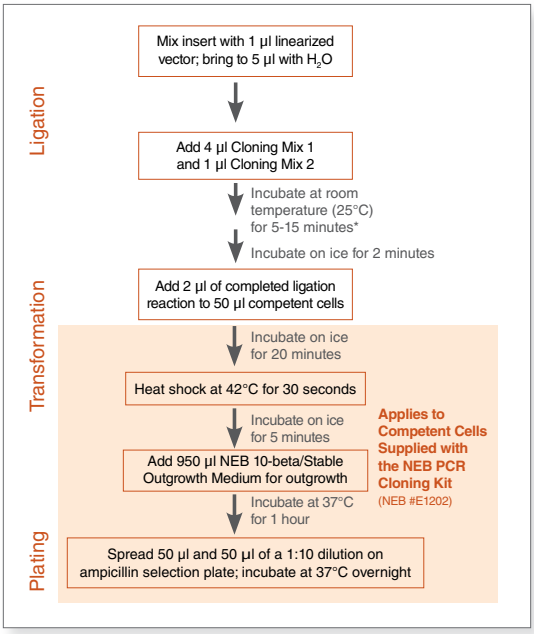
★★★ Optimal, recommended product for selected application
★★ Works well for selected application
★ Will perform selected application, but is not recommended
N/A Not applicable to this application
NR Not recommended

NEB offers several online tools to aid in DNA assembly. Visit the Tools & Resources tab at www.neb.com for the full list.

NEB® PCR Cloning Kit

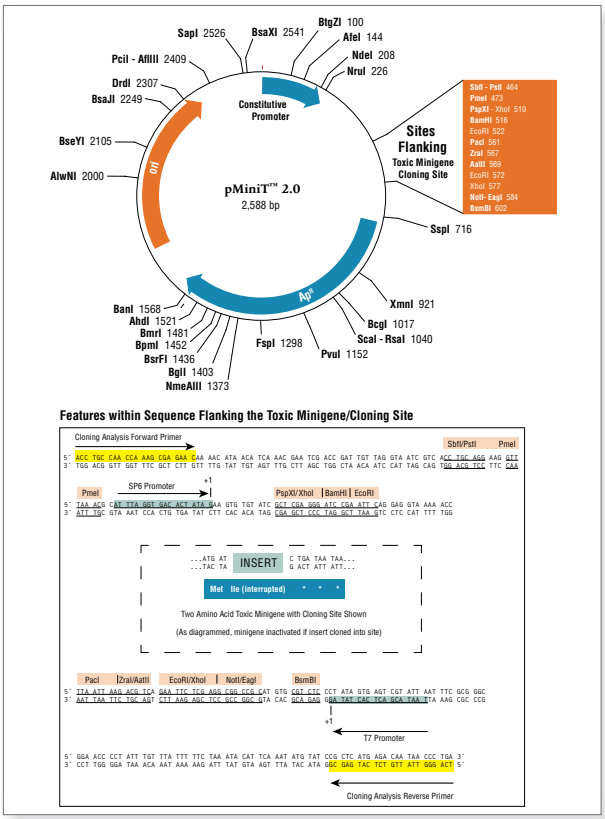
The NEB PCR Cloning Kit enables quick and simple cloning of all your PCR amplicons, regardless of the polymerase used. This kit utilizes a novel mechanism for background colony suppression – a toxic minigene is generated when the vector closes upon itself – and allows for direct cloning from your reaction, with no purification step.

Cloning Kit Protocol Overview



*Note: While 5 minutes is recommended, 15 minutes will increase transformation levels for inserts suspected as being difficult to clone.

Vector Maps and Sequence



Map shown displays the construct formed if no insert is present. Unique restriction sites are shown in bold. Additional restriction sites that can be used for subcloning are also shown. Expanded box below shows location of sequencing primers, restriction sites for subcloning or linearization for in vitro transcription, RNA Polymerase promoter sequences and placement of insertion site within the toxic minigene.

RECOMMENDED PRODUCTS

NEB PCR Cloning Kit (NEB #E1202)

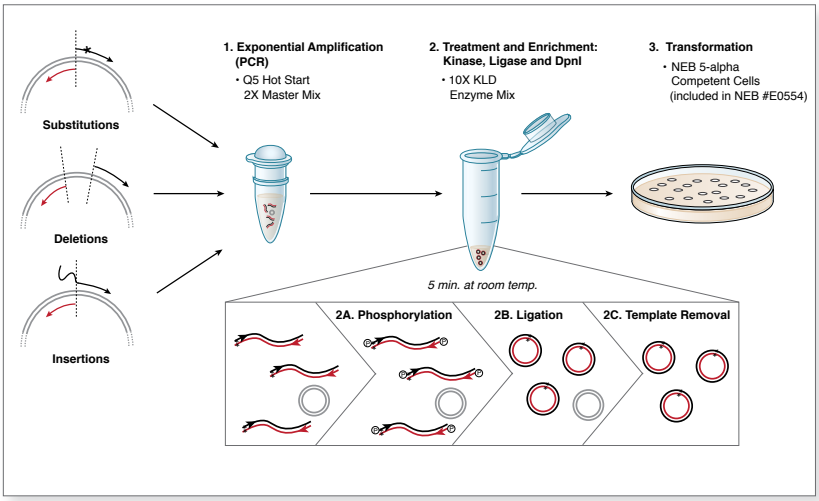
NEB PCR Cloning Kit (without competent cells) (NEB #E1203)

- Easy cloning of all PCR products, including blunt and TA ends
- Fast cloning experiments with 5-minute ligation step
- Simplified screening with low/no colony background and no blue/white selection required
- Save time by eliminating purification steps
- Updated to allow for *in vitro* transcription with both SP6 and T7 promoters
- Flanking restriction sites available for easy subcloning, including choice of two single digest options
- Provided analysis primers allow for downstream colony PCR screening or sequencing
- Ready-to-use kit components include 1 kb control amplicon, linearized cloning vector and optional single-use competent *E. coli*
- Longer shelf life (12 months), as compared to some commercially available products
- Value pricing

Q5® Site-Directed Mutagenesis Kit

The Q5 Site-Directed Mutagenesis Kit enables rapid, site-specific mutagenesis of double-stranded plasmid DNA in less than 2 hours. The kit utilizes Q5 Hot Start High-Fidelity DNA Polymerase, along with custom mutagenic primers to create substitutions, deletions and insertions in your vector of choice. Transformation into high-efficiency NEB 5-alpha Competent *E. coli* cells ensures robust results with plasmids up to 14.3 kb in length.

Overview of Q5 Site-Directed Mutagenesis Kit



RECOMMENDED PRODUCTS

Q5 Site-Directed Mutagenesis Kit (NEB #E0554)

Q5 Site-Directed Mutagenesis Kit (Without Competent Cells) (NEB #E0552)

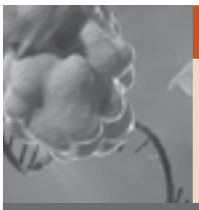
KLD Enzyme Mix (NEB #M0554S)

- Generation of mutations, insertions or deletions in plasmid DNA
- Non-overlapping primer design ensures robust, exponential amplification and generates a high % of desired mutations
- Low error rate of Q5 High-Fidelity DNA Polymerase reduces screening time
- Use of standard primers eliminates need for phosphorylated or purified oligos
- Easy-to-use master mix format
- For help with primer design, try NEBaseChanger™ at NEBaseChanger.neb.com

Amplify your confidence with our PCR polymerases

Whether you are a seasoned cloner or a first-timer, choosing the optimal PCR polymerase for your target amplicon is essential. With NEB's selection of polymerases for standard, high-fidelity and specialty PCR, you can have confidence in the reliability of amplification, regardless of sequence composition. For example, our high-fidelity polymerases ensure accuracy in the amplification of your insert. However, when fidelity is less important, such as for screening or colony PCR, you'll get robust yields with minimal optimization using one of our routine polymerases, all at a lower price. Add flexibility to your reaction setup with enzyme, kit or master mix formats. Let NEB's PCR Polymerases amplify your cloning confidence!

PCR Polymerase Selection Chart for Cloning



	STANDARD PCR		HIGH-FIDELITY PCR		SPECIALTY PCR
			Highest Fidelity		Long Amplicons
	One *Taq*/ One *Taq* Hot Start	*Taq* / Hot Start *Taq*	Q5/ Q5 Hot Start	Phusion®(1)/ Phusion(1) Flex	LongAmp®/ LongAmp Hot Start *Taq*
PROPERTIES					
Fidelity vs. *Taq*	2X	1X	~280X(3)	> 50X	2X
Amplicon Size	< 6 kb	≤ 5 kb	≤ 20 kb	≤ 20 kb	≤ 30 kb
Extension Time	1 kb/min.	1 kb/min.	6 kb/min.	4 kb/min.	1.2 kb/min.
Resulting Ends	3´ A/Blunt	3´ A	Blunt	Blunt	3´ A/Blunt
3´→ 5´ exo	Yes	No	Yes	Yes	Yes
5´→ 3´ exo	Yes	Yes	No	No	Yes
Units/50 µl Reaction	1.25	1.25	1.0	1.0	5.0
Annealing Temperature	Tm¯5	Tm¯5	Tm⁺3	Tm⁺3	Tm¯5
APPLICATIONS					
Routine PCR	★	●	●	●	●
Colony PCR	★	●			●
Enhanced Fidelity	●		★	●	●
High Fidelity			★	●	
High Yield	★	●	★	●	
Fast			★	●	
Long Amplicon			★	●	★
GC-rich Targets	★		★		●
AT-rich Targets	★	●	★	●	●
High Throughput	●	●	●	●	
Multiplex PCR	●	★(2)	●	●	
Site-directed Mutagenesis			★	●	
FORMATS					
Hot Start Available	●	●	●	●	●
Kit		●	●	●	●
Master Mix Available	●	●	●	●	●
Direct Gel Loading	●	●			

(1) Phusion DNA Polymerase was developed by Finnzymes Oy, now a part of Thermo Fisher Scientific. This product is manufactured by New England Biolabs, Inc. under agreement with, and under the performance specifications of Thermo Fisher Scientific.

(2) Use Multiplex PCR 5X Master Mix.

(3) We continue to investigate improved assays to characterize Q5's very low error rate to ensure that we present the most accurate fidelity data possible (Potapov, V. and Ong, J.L. (2017) PLoS ONE. 12(1): e0169774.)

★ indicates recommended choice for application

RECOMMENDED PRODUCTS

Q5 High-Fidelity DNA Polymerase

- ~280X the fidelity of *Taq* DNA Polymerase
- High specificity and robust yields with minimal optimization
- Superior performance for a broad range of amplicons (from high AT to high GC)
- Fast, with short extension times (10–30 s per kb)

One*Taq*® DNA Polymerase

- Exceptional performance across a wide range of templates (from high AT to high GC)
- Hot Start version allows room temperature reaction setup; no activation step
- Compatible with standard *Taq* protocols

TOOLS & RESOURCES

Visit NEBPCRPolymerases.com to find:

- The full list of polymerases available
- FAQs & troubleshooting guides
- Interactive tools to help with experimental design
- Videos for setting up PCR reactions

Did you know that most high-fidelity polymerases benefit from a Tm⁺3 annealing temperature? Use the **NEB Tm Calculator** at www.neb.com/TmCalculator.com to ensure successful PCR.

Ensure robust, full length cDNA synthesis – every time

Of the many steps in the cloning workflow, cDNA synthesis is one of the more complex. To facilitate the generation of high-quality, full-length cDNAs, NEB offers a selection of reverse transcriptases (RTs), which are available as standalone enzymes or kits, to enable workflow customization. With RNA as a starting material, there are a number of important considerations. For difficult targets or those with complex secondary structure, you'll want to choose an RT with high thermostability, as thermostable RTs are able to perform at higher temperatures on relaxed secondary structures. RTs with RNase H activity can degrade RNA:DNA hybrids, negatively impacting cDNA length and yield. Choosing an RT with reduced RNase H activity (RNase H^-) increases yield, and results in more full-length cDNA product. Following reverse transcription, we recommend using a high-fidelity DNA polymerase to generate double-stranded (ds) cDNA for cloning.

RECOMMENDED PRODUCT

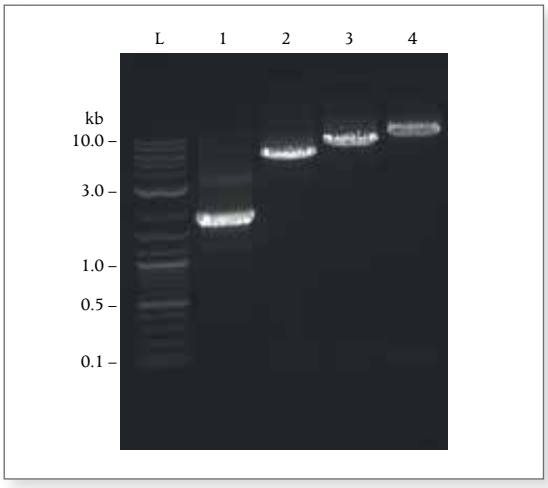
ProtoScript® II First Strand cDNA Synthesis Kit (NEB #E6560)

- High-quality, robust cDNA synthesis for a wide range of templates
- Master mix formulation speeds reaction setup
- Improved thermostability over standard M-MuLV Reverse Transcriptase
- Increased performance and value
- Also available as a standalone enzyme

cDNA Synthesis Selection Chart

cDNA SYNTHESIS	FEATURES
KIT	
ProtoScript First Strand cDNA Synthesis Kit (NEB #E6300)	Generates cDNA at least 5 kb in length
	Contains M-MuLV Reverse Transcriptase
	Convenient 2-tube kit includes dNTPs, Oligo-dT primer and Random Primer Mix
ProtoScript® II First Strand cDNA Synthesis Kit (#E6560S/L)	Convenient 2-tube kit
	Contains ProtoScript II Reverse Transcriptase
	Increased thermostability as compared to M-MuLV
	Reduced RNase H activity
	Generates cDNA at least 10 kb in length
	Includes dNTPs, Oligo-dT primer and Random Primer Mmix
STANDALONE REAGENT	
ProtoScript II Reverse Transcriptase (NEB #M0368) *An alternative to SuperScript® II*	RNase H^- mutant of M-MuLV Reverse Transcriptase with increased thermostability and reduced RNase H activity
	Increased reaction temperatures (37–50°C)
M-MuLV Reverse Transcriptase (NEB #M0253)	Robust reverse transcriptase for a variety of templates
	Standard reaction temperatures (37–45°C)
AMV Reverse Transcriptase (NEB #M0277)	Robust reverse transcriptase for a broad temperature range (37–52°C)
	Can be used for templates requiring higher reaction temperatures

cDNA synthesis for RT-PCR up to 10 kb using the ProtoScript II First Strand cDNA Synthesis Kit

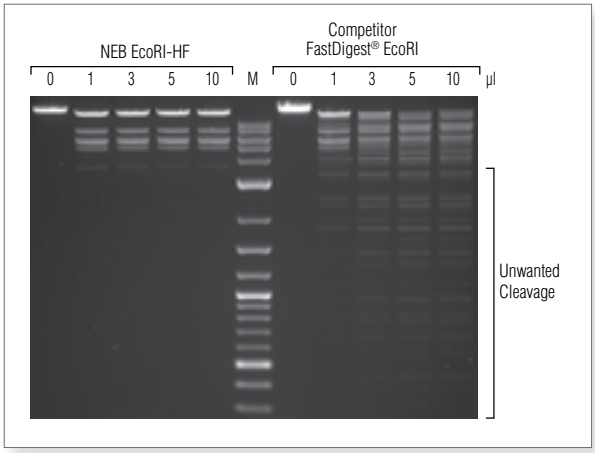


Jurkat total RNA (250 ng) was converted to first strand cDNA using the ProtoScript II First Strand cDNA Synthesis Kit. (For more details see page 2)

Experience the convenience of NEB restriction enzymes

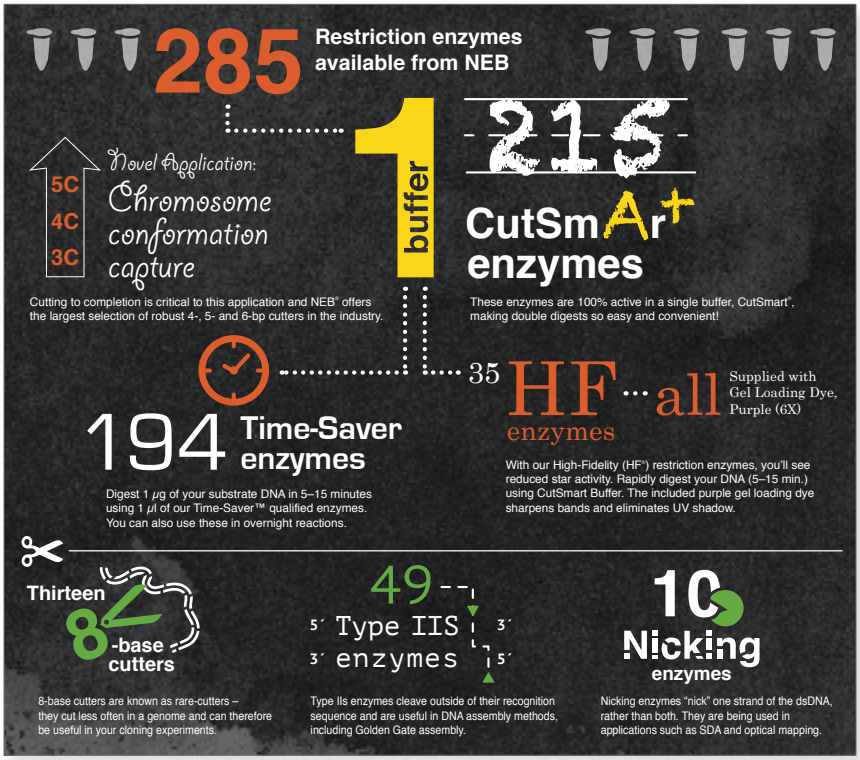
Let NEB restriction enzymes bring convenience to your cloning workflow. With over 210 restriction enzymes that are 100% active in a single buffer, CutSmart™ Buffer, it is much simpler to set up your double digest reactions. Furthermore, you can speed up your reactions by choosing one of our Time-Saver™ qualified restriction enzymes. These enzymes will digest DNA in 5–15 minutes, and can also be used safely overnight with no loss of sample. With over 230 specificities to choose from, you can be sure to find the enzyme you need.

For the full list of restriction enzymes available from NEB, as well as the latest activity/performance chart, visit **NEBRestrictionEnzymes.com**.



EcoRI-HF (NEB #R3101) shows no star activity in overnight digests, even when used at higher concentrations. 50 µl reactions were set up using 1 µg of Lambda DNA, the indicated amount of enzyme and the recommended reaction buffer. Reactions were incubated overnight at 37°C. Marker M is the 1 kb DNA Ladder (NEB# N3232).

NEB Restriction Enzymes by the Numbers



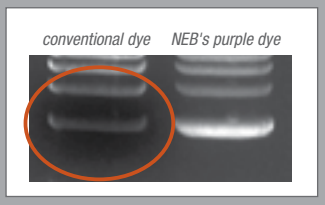
RECOMMENDED PRODUCTS

High-Fidelity (HF®) Restriction Enzymes

- One-buffer convenience with no loss of performance (CutSmart Buffer)
- Reduced star activity eliminates unwanted cleavage
- Time-Saver qualified for 5–15 minute digests or can digest safely overnight without sample loss
- Engineered performance under a wide range of conditions
- Added flexibility without added cost
- 6X Gel-Loading Dye, Purple included with most restriction enzymes and all HFs –sharpens bands and leaves no UV shadow

Gel Loading Dye, Purple (6X) (NEB #B7024)

- Brighter, sharper bands vs. glycerol based dyes
- No UV shadow, allowing for publication grade images



No UV-shadow and sharp bands: **Lane 1:** UV shadow typical for conventional loading dyes (e.g. Bromophenol-Blue); **Lane 2:** NEB's purple loading buffer as supplied with all top selling restriction enzymes

COMPARISON OF DYE FRONTS

TOOLS & RESOURCES

Visit NEBRestrictionEnzymes.com to find:

- The full list of restriction enzymes available
- The latest activity/performance chart
- Videos for setting up restriction enzyme digests, double digestions and troubleshooting reactions

Online interactive tools to help with setting up your restriction enzyme digests:

- Enzyme Finder
- Double Digest Finder
- NEBcutter®
- NEBioCalculator®
- REBASE®
- NEBcloner

DNA end modifications that will improve your results

Make blunting fast and easy

Deciding which blunting enzyme to use depends on which activity you are looking for: fill in of a 5´ overhang, removal of a 3´ overhang or removal of a 5´ overhang. Once you have blunted your vector, a dephosphorylation step may still be necessary to prevent self-ligation.

Blunting Selection Chart

	T4 DNA Polymerase* (NEB #M0203)	DNA Polymerase I, Large (Klenow) Fragment (NEB #M0210)	Quick Blunting Kit (NEB #E1201)	Mung Bean Nuclease (NEB #M0250)
APPLICATION				
Fill in of 5´ overhangs	●	●	●	
Removal of 3´ overhangs	●	●	●	●
Removal of 5´ overhangs				●

* T4 DNA Polymerase has a strong 3´→ 5´ exo activity.

RECOMMENDED PRODUCT

Quick Blunting™ Kit (NEB #E1201)

- Optimized mix for blunting up to 5 µg of DNA in less than 30 minutes
- Reactions are performed at room temperature in a ready-to-use mix
- Contains T4 DNA Polymerase and T4 Polynucleotide Kinase, enabling blunting and phosphorylation in one step

BLUNTING WITH DNA POLYMERASES

Reduce background with dephosphorylation

Did you know that it's possible to reduce background from your cloning vector by dephosphorylating your vector prior to ligation? A heat-labile phosphatase with high specific activity is the best choice for dephosphorylation. This allows you to add the enzyme directly to most buffers, quickly heat inactivate, and move on to the next step in your workflow. No need to purify.

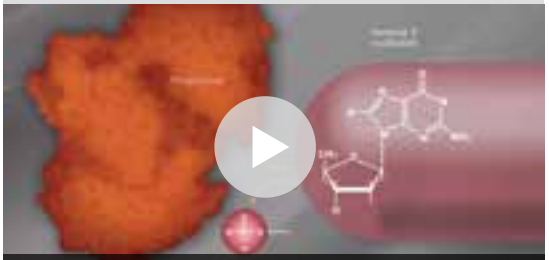
Phosphatases Selection Chart

	NEW QUICK DEPHOSPHORYLATION KIT	RECOMBINANT SHRIMP ALKALINE PHOSPHATASE (rSAP)	ANTARCTIC PHOSPHATASE	ALKALINE PHOSPHATASE CALF INTESTINAL (CIP)
FEATURES				
100% heat inactivation	2 minutes/80°C	5 minutes/65°C	2 minutes/80°C	No
High specific activity	●	●		●
Improved stability	●	●		
Works directly in NEBuffers	●	●	●	●
Requires additive			● (Zn^{2+})	
Quick Protocol	●			

RECOMMENDED PRODUCT

Quick Dephosphorylation Kit (NEB #M0508)

- Complete dephosphorylation in 10 minutes
- Rapid and irreversible heat inactivation eliminates unwanted activity
- Faster reaction setup (no supplemental additives like zinc required)
- Flexible reaction conditions (active in any restriction enzyme buffer, no clean-up required)
- No need for multiple phosphatases (removes 5´- and 3´- phosphates from DNA, RNA and dNTPs)
- Active on unincorporated dNTPs in PCR products – improves DNA sequencing and SNP analysis



THE MECHANISM OF DEPHOSPHORYLATION

Easily generate dA-tails for PCR cloning

Tailing is typically done to prepare a T-vector for use in TA cloning or to dA-tail a PCR product produced by a high-fidelity polymerase (not *Taq* DNA Polymerase) for use in TA cloning.

Tailing Selection Chart

	Klenow Fragment (3´→5´ exo-) (NEB #M0212)	*Taq* DNA Polymerase
FEATURES		
Reaction temperature	37°C	75°C
Heat inactivated	75°C, 20 minutes	No
Nucleotide cofactor	dATP	dATP

RECOMMENDED PRODUCT

Klenow Fragment (3´→5´ exo-) (NEB #M0212)

- Fully active in NEBuffers 1.1, 2.1, 3.1 and CutSmart (when supplemented with dNTPs), which eliminates the need to clean up the reaction for downstream applications
- Heat inactivated at 75° C after 20 minutes

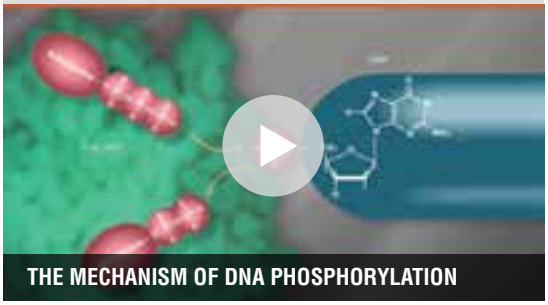
When is a phosphorylation step needed?

Typically, PCR primers are not phosphorylated; the 5´ ends of the amplicon need to be treated by a kinase to introduce the 5´ phosphate necessary for ligation, if cloning into a dephosphorylated vector.

RECOMMENDED PRODUCT

T4 Polynucleotide Kinase (NEB #M0201)

- Highly-efficient catalysis of the 5´addition of P_i to ds- & ssDNA, RNA, as well as nucleoside 3´monophosphates



Featured Online Tool: NEBCloner

Use NEBcloner at **NEBcloner.neb.com** to find the right products and protocols for each step in your traditional cloning experiment. Also find other relevent tools and resources to enable protocol optimization.

Digestion

View a protocol for cutting DNA with a single restriction enzyme or a pair of restriction enzymes.

End Modification

View recommended products and protocols for blunting, dephosphorylation, phosphorylation, and A-tailing of DNA ends.

Ligation

View recommended products and protocols for joining DNA ends based on your input parameters.

Transformation

Select competent cells and view transformation protocols based on your input parameters.

Easy & effective ligation for all end types

Once your insert and vector are end-treated, as needed, and have compatible ends, you'll be ready to ligate. NEB offers an extensive selection of high-quality and performance-optimized DNA ligases and ligase master mixes; you'll be certain to find an enzyme specific for your end type, enabling maximum efficiency. Since time is always at a premium in the lab, you can speed up your reaction setup and incubation by choosing one of our ligase master mixes. With so many ligases to choose from, make NEB your source for DNA ligases.

DNA Ligase Selection Chart for Cloning

	Instant Sticky-end Ligase Master Mix (NEB #M0370)	Blunt/TA Ligase Master Mix (NEB #M0367)	ElectroLigase® (NEB #M0369)	T4 DNA Ligase (NEB #M0202)	Quick Ligation Kit (NEB #M2200)	T3 DNA Ligase (NEB #M0317)	T7 DNA Ligase (NEB #M0318)	*Taq* DNA Ligase (NEB #M0208)
DNA APPLICATIONS								
Ligation of sticky ends	●●●	●●	●●	●●	●●●	●●	●●	●
Ligation of blunt ends	●	●●●	●●	●●	●●●	●●		
T/A cloning	●	●●●	●●	●●	●●	●	●	
Electroporation			●●●	●●				
Ligation of sticky ends only							●●●	
Repair of nicks in dsDNA	●●●	●●●	●●●	●●●	●●●	●●●	●●●	●●●
High complexity library cloning	●●	●●	●●	●●●	●●			
FEATURES								
Salt tolerance (> 2X that of T4 DNA Ligase)						✔		
Ligation in 15 min. or less	✔	✔		✔	✔	✔	✔	✔
Master Mix Formulation	✔	✔						
Thermostable								✔
Recombinant	✔	✔	✔	✔	✔	✔	✔	✔

KEY

- ●●● Recommended ligase for selected application
- ●● Works well for selected application
- ● Will perform selected application, but is not recommended

RECOMMENDED PRODUCTS

- For the ligation of sticky ends, NEB recommends our **Instant Sticky-end Ligase Master Mix (NEB #M0370)** (no incubation needed) or the **Quick Ligation™ Kit (NEB #M2200)**
- For the ligation of blunt ends, NEB recommends our **Blunt/TA Ligase Master Mix (NEB #M0367)** or the **Quick Ligation Kit (NEB #M2200)**
- The **Blunt/TA Ligase Master Mix (NEB #M0367)** is recommended for T/A cloning

TOOLS & RESOURCES

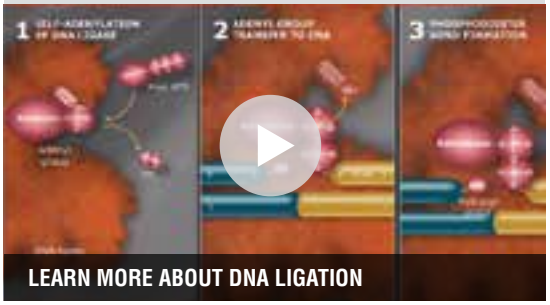
Visit NEBStickTogether.com to find:

- The full list of ligases available
- FAQs
- Videos about ligation and help with setting up ligation reactions

Online interactive tools to help with setting up your restriction enzyme digests:

NEBcloner

NEBioCalculator



Maximize your transformation efficiency

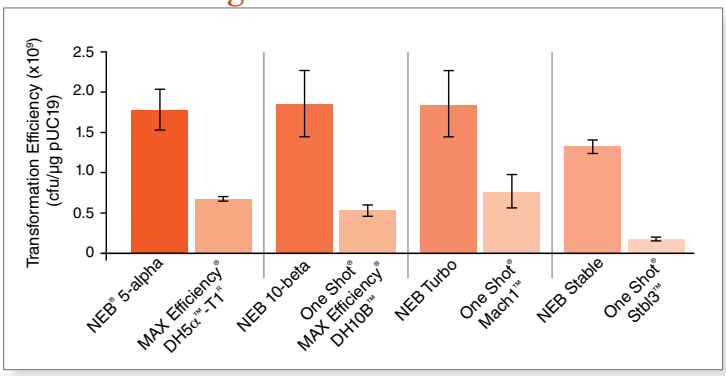
Selecting a competent cell line for cloning requires that you strike the right balance; you want a strain that offers high transformation efficiencies, but also has the ability to generate sufficient plasmid yields. NEB cloning strains offer high transformation efficiencies, and are T1 phage resistant and endA deficient, ensuring high-quality plasmid preparation. Additionally, RecA⁻ strains minimize potential plasmid recombination. For convenience, NEB cloning strains are available in a variety of formats, including single-use tubes, 96-well plate, 384-well plate, and strip tubes. Electrocompetent and subcloning efficiencies are also available. Your choice of competent cell line for cloning can influence the success of your cloning experiment; NEB offers a cell line for every need.

Competent Cell Selection Chart

	NEB 5-alpha Competent *E. coli* (NEB #C2987)	NEB Turbo Competent *E. coli* (NEB #C2984)	NEB 5-alpha F´ I^q Competent *E. coli* (NEB #C2992)	NEB 10-beta Competent *E. coli* (NEB #C3019)	*dam*⁻/*dcm*⁻ Competent *E. coli* (NEB #C2925)	NEB Stable Competent *E. coli* (NEB #C3040)
FEATURES						
Versatile	•			•		•
Fast growth (< 8 hours)		•				
Toxic gene cloning		•	•			•
Large plasmid/BAC cloning				•		•
Dam/Dcm free plasmid growth					•	
Retroviral/lentiviral vector cloning						•
RecA-	•		•	•		•
FORMATS						
Chemically Competent	•	•	•	•	•	•
Electrocompetent	•	•		•		
Subcloning	•					
96-well format*	•					
384-well format*	•					
12 x 8 tube strips*	•					

**Other strains are available on request. for more information contact custom@neb.com.*

Benefit from High Transformation Efficiencies



The transformation efficiencies were compared using manufacturers' recommended protocols. Values shown are the average of triplicate experiments.

RECOMMENDED PRODUCTS

NEB 5-alpha Competent *E.coli* (NEB #C2987)

- Cloning strain with same genetic features as the popular DH5α™
- High transformation efficiencies, available as electrocompetent or chemically competent
- Convenient formats, including single-use tubes, 96-well plate, 384-well plate, and strip tubes
- Value pricing
- RecA⁻ strain

TOOLS & RESOURCES

Visit www.neb.com/CloningCompCells to find:

- The full list of competent cells available
- FAQs
- Access to our **Competitor Cross-Reference Tool**, for finding the NEB strain that is compatible to other commercially available strains
- Technical tips and troubleshooting guides
- Protocol Videos

THE MECHANISM OF TRANSFORMATION WITH COMPETENT CELLS

NO DRY ICE CHARGES
with Competent Cells from NEB

Take DNA analysis to the next level

NEB's DNA ladders help to take the guesswork out of your sample analysis during gel electrophoresis. With sharp, evenly spaced reference bands that are easy to identify, our DNA Ladders can be used for sample quantitation. Choose the DNA ladder that best suits your target insert and vector sizes. Try our Quick-Load Purple DNA Ladders, which include a vial of our Purple Gel Loading Dye (no SDS), for sharper bands and no UV shadow.

RECOMMENDED PRODUCTS

Quick-Load® Purple 2-Log DNA Ladder (NEB #N0550)

Quick-Load Purple 100 bp DNA Ladder (NEB #N0551)

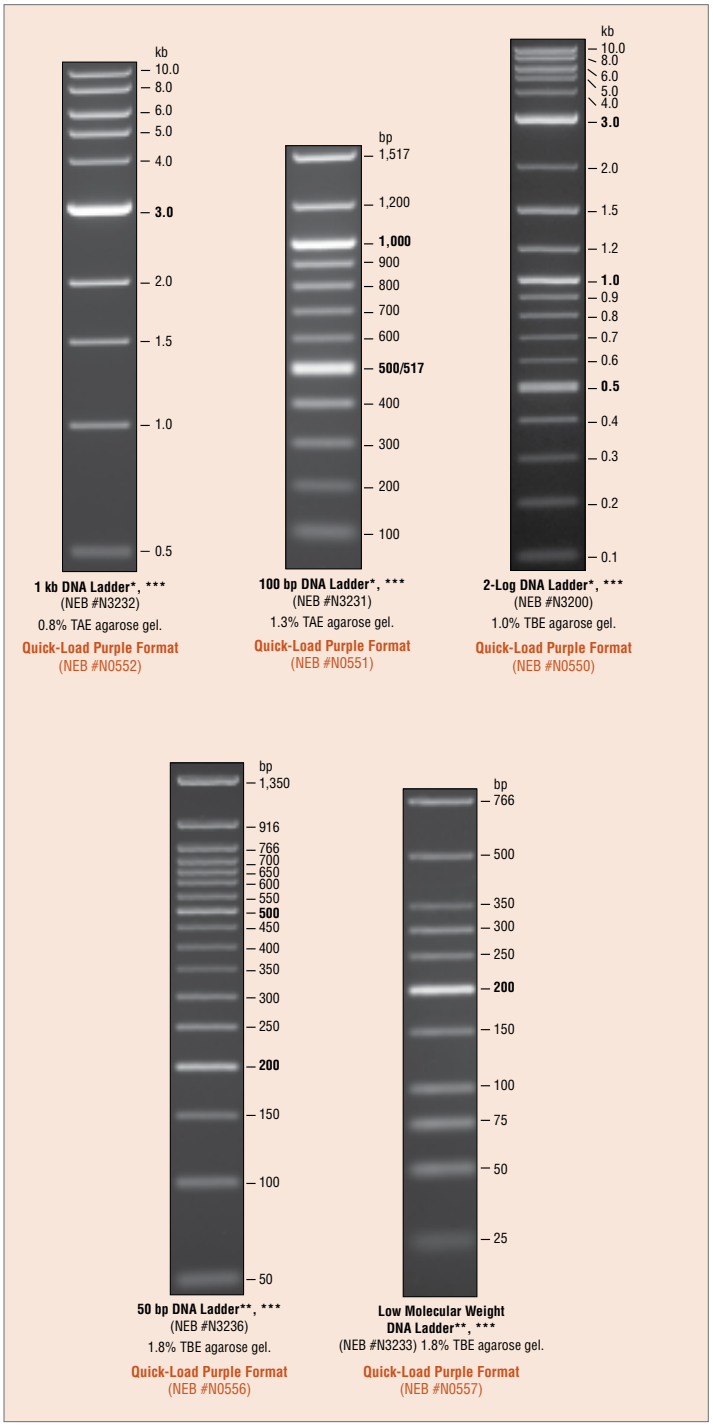
Quick-Load Purple 1 kb DNA Ladder (NEB #N0552)

Quick-Load Purple 50 bp DNA Ladder (NEB #N0556)

Quick-Load Purple Low Molecular Weight DNA Ladder (NEB #N0557)

- Ideal for analysis from 0.1–10 kb
- Convenient, ready-to-load format
- Sharp, high contrast bands
- Utilizes purple gel loading dye which is also supplied with HF restriction enzymes
- Includes vial of purple dye (no SDS)

DNA Ladders Available in Quick-Load Purple Format



1 kb DNA Ladder*, *** (NEB #N3232)
0.8% TAE agarose gel.
Quick-Load Purple Format (NEB #N0552)

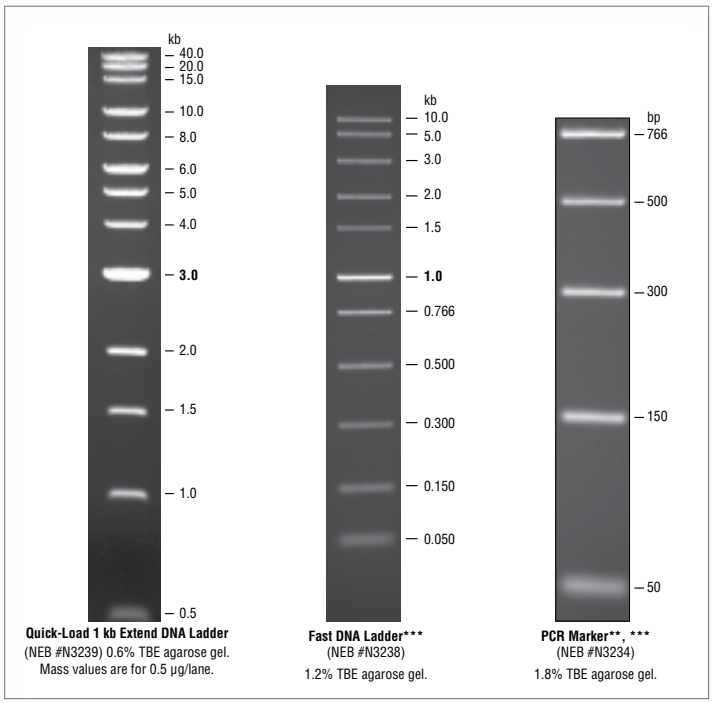
100 bp DNA Ladder*, *** (NEB #N3231)
1.3% TAE agarose gel.
Quick-Load Purple Format (NEB #N0551)

2-Log DNA Ladder*, *** (NEB #N3200)
1.0% TBE agarose gel.
Quick-Load Purple Format (NEB #N0550)

50 bp DNA Ladder, ***** (NEB #N3236)
1.8% TBE agarose gel.
Quick-Load Purple Format (NEB #N0556)

Low Molecular Weight DNA Ladder, *****
(NEB #N3233) 1.8% TBE agarose gel.
Quick-Load Purple Format (NEB #N0557)

Additional DNA Ladders from New England Biolabs



Quick-Load 1 kb Extend DNA Ladder
(NEB #N3239) 0.6% TBE agarose gel.
Mass values are for 0.5 µg/lane.

Fast DNA Ladder***
(NEB #N3238)
1.2% TBE agarose gel.

PCR Marker, *****
(NEB #N3234)
1.8% TBE agarose gel.

** Available in Quick-Load® and TriDye™ formats ** Available in Quick-Load format *** Free Loading Dye included*

Monarch® Nucleic Acid Purification Kits

Monarch kits provide fast and reliable purification of high quality DNA from bacterial cultures, agarose gels, and enzymatic reactions using best-in-class technology. Our unique column design offers elution in lower volumes than standard purification kits, providing concentrated, high quality DNA suitable for use in downstream applications such as DNA sequencing, PCR, restriction enzyme digests and other enzymatic manipulations. Our column design also eliminates buffer retention and the risk of carryover contamination, providing fast, worry-free DNA purification. Designed with sustainability in mind, Monarch kits use significantly less plastic and responsibly-sourced packaging.

Designed for performance

Many purification columns are built with a frit to hold the membrane in place. This frit can trap buffer during various steps in the protocol. Monarch Columns' silica matrix is held in place without the use of a frit, thereby eliminating buffer retention and ensuring worry-free purification.

RECOMMENDED PRODUCTS

Monarch Plasmid Miniprep Kit (NEB #T1010)

Monarch DNA Gel Extraction Kit (NEB #T1020)

Monarch PCR & DNA Cleanup Kit (NEB #T1030)

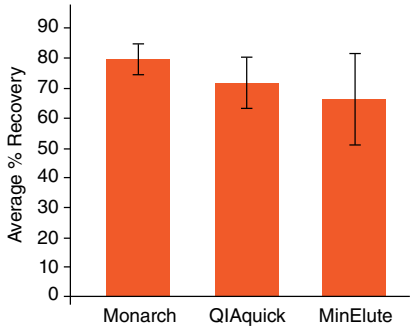
- Optimized for maximum performance and minimal environmental impact
- Unique column design eliminates buffer retention and offers elution in lower volumes
- Fast, user-friendly protocols

TOOLS & RESOURCES

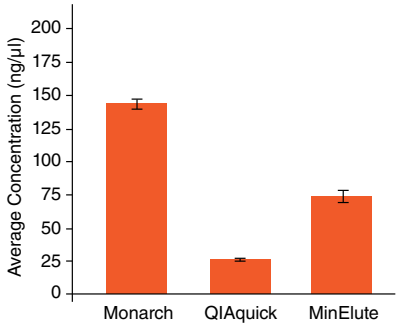
Visit NEBMonarch.com to find:

- Protocol videos and optimization tips
- Troubleshooting guide & FAQs
- Tips for recycling your Monarch kit

DNA purified from the Monarch DNA Gel Extraction Kit is recovered with similar efficiency and purity as the leading supplier, but is more highly concentrated, facilitating its use in downstream applications



	Monarch	QIAquick®	MinElute®
$A_{260/280}$	1.9+/-0.08	1.9+/-0.08	1.9+/-0.04
Yield (µg)	0.80+/-0.05	0.72+/-0.09	0.66+/-0.13



	Monarch	QIAquick®	MinElute®
Conc. (ng/µl)	144.9+/-9.6	25.7+/-3.1	73.8+/-14.2

One microgram aliquots of a 3 kb fragment were resolved on a 1% w/v agarose gel, excised, and processed with different kits using manufacturer-specified minimum elution volumes. Values reported are the concentration and purity data determined by Nanodrop™ readings, as well as recovery calculations based on the eluted DNA concentration and recovered volume.

ORDERING INFORMATION

Selected Products for PCR

PRODUCT	NEB #	SIZE
HIGH-FIDELITY DNA POLYMERASES		
Q5 High-Fidelity DNA Polymerase	M0491S/L	100/500 units
Q5 Hot Start High-Fidelity DNA Polymerase	M0493S/L	100/500 units
Q5 High-Fidelity 2X Master Mix	M0492S/L	100/500 reactions
Q5 Hot Start High-Fidelity 2X Master Mix	M0494S/L	100/500 reactions
Q5 High-Fidelity PCR Kit	E0555S/L	50/200 reactions
Phusion High-Fidelity PCR Master Mix with HF Buffer	M0531S/L	100/500 reactions
Phusion High-Fidelity PCR Master Mix with GC Buffer	M0532S/L	100/500 reactions
Phusion Hot Start Flex 2X Master Mix	M0536S/L	100/500 reactions
Phusion High-Fidelity PCR Kit	E0553S/L	50/200 reactions
Phusion High-Fidelity DNA Polymerase	M0530S/L	100/500 units
Phusion Hot Start Flex High-Fidelity DNA Polymerase	M0535S/L	100/500 units
DNA POLYMERASES		
One*Taq* DNA Polymerase	M0480S/L/X	200/1,000/5,000 units
One*Taq* Hot Start DNA Polymerase	M0481S/L/X	200/1,000/5,000 units
One*Taq* 2X Master Mix with Standard Buffer	M0482S/L	100/500 reactions
One*Taq* 2X Master Mix with GC Buffer	M0483S/L	100/500 reactions
One*Taq* Quick-Load 2X Master Mix with GC Buffer	M0487S/L	100/500 reactions
One*Taq* Quick-Load 2X Master Mix with Standard Buffer	M0486S/L	100/500 reactions
One*Taq* Hot Start 2X Master Mix with Standard Buffer	M0484S/L	100/500 reactions
One*Taq* Hot Start 2X Master Mix with GC Buffer	M0485S/L	100/500 reactions
One*Taq* Hot Start Quick-Load 2X Master Mix with Standard Buffer	M0488S/L	100/500 reactions
One*Taq* Hot Start Quick-Load 2X Master Mix with GC Buffer	M0489S/L	100/500 reactions
Taq DNA Polymerase with ThermoPol™ Buffer	M0267S/L/X/E	400/2,000/4,000/20,000 units
Taq DNA Polymerase with ThermoPol II (Mg-free) Buffer	M0321S/L	400/2,000 units
Taq DNA Polymerase with Standard *Taq* Buffer	M0273S/L/X/E	400/2,000/4,000/20,000 units
Taq DNA Polymerase with Standard *Taq* (Mg-free) Buffer	M0320S/L	400/2,000 units
Taq PCR Kit	E5000S	200 reactions
Quick-Load *Taq* 2X Master Mix	M0271L	500 reactions
Taq 2X Master Mix	M0270L	500 reactions
Taq 5X Master Mix	M0285L	500 reactions
Multiplex PCR 5X Master Mix	M0284S	100 reactions
Hot Start *Taq* DNA Polymerase	M0495S/L	200/1,000 units
Hot Start *Taq* 2X Master Mix	M0496S/L	100/500 reactions
LongAmp *Taq* DNA Polymerase	M0323S/L	500/2,500 units
LongAmp Hot Start *Taq* DNA Polymerase	M0534S/L	500/2,500 units
LongAmp *Taq* 2X Master Mix	M0287S/L	100/500 reactions
LongAmp Hot Start *Taq* 2X Master Mix	M0533S/L	100/500 reactions
LongAmp *Taq* PCR Kit	E5200S	100 reactions
PCR CLONING & MUTAGENESIS		
NEB PCR Cloning Kit	E1202S	20 reactions
NEB PCR Cloning Kit (Without Competent Cells)	E1203S	20 reactions
Q5 Site-Directed Mutagenesis Kit	E0554S	10 reactions
Q5 Site-Directed Mutagenesis Kit (Without Competent Cells)	E0552S	10 reactions
KLD Enzyme Mix	M0554S	25 reactions

Selected Products for PCR (Cont.)

PRODUCT	NEB #	SIZE
dNTPs		
Deoxynucleotide (dNTP) Solution Set	N0446S	25 µmol of each
Deoxynucleotide (dNTP) Solution Mix	N0447S/L	8/40 µmol of each

Products for cDNA Synthesis

PRODUCT	NEB #	SIZE
ProtoScript II First Strand cDNA Synthesis Kit	E6560S/L	30/150 reactions
ProtoScript First Strand cDNA Synthesis Kit	E6300S/L	30/150 reactions
ProtoScript II Reverse Transcriptase	M0368S/L/X	4,000/10,000/40,000 units
M-MuLV Reverse Transcriptase	M0253S/L	10,000/50,000 units
AMV Reverse Transcriptase	M0277S/L	200/1,000 units
WarmStart RTx Reverse Transcriptase	M0380S/L	50/250 reactions

Products for Restriction Digestion

PRODUCT	NEB #	SIZE
HIGH-FIDELITY (HF®) RESTRICTION ENZYMES		
AgeI-HF	R3552S/L	300/1,500 units
ApoI-HF	R3566S/L	1,000/5,000 units
BamHI-HF	R3136S/L/T/M	10,000/50,000 units
BbsI-HF	R3539S/L	300/1,500 units
BclI-HF	R3160S/L	3,000/15,000 units
BmtI-HF	R3658S/L	300/1,500 units
BsaI-HF	R3535S/L	1,000/5,000 units
BsiWI-HF	R3553S/L	300/1,500 units
BsrFαI-HF	R0682S/L	1,000/5,000 units
BsrGI-HF	R3575S/L	1,000/5,000 units
BstEII-HF	R3162S/L/M	2,000/10,000 units
BstZ17I-HF	R3594S/L	1,000/5,000 units
DraIII-HF	R3510S/L	1,000/5,000 units
EagI-HF	R3505S/L/M	500/2,500 units
EcoRI-HF	R3101S/L/T/M	10,000/50,000 units
EcoRV-HF	R3195S/L/T/M	4,000/20,000 units
HindIII-HF	R3104S/L/T/M	10,000/50,000 units
KpnI-HF	R3142S/L/M	4,000/20,000 units
MfeI-HF	R3589S/L	500/2,500 units
MluI-HF	R3198S/L	1,000/5,000 units
NcoI-HF	R3193S/L/M	1,000/5,000 units
NheI-HF	R3131S/L/M	1,000/5,000 units
NotI-HF	R3189S/L/M	500/2,500 units
NruI-HF	R3192S/L	1,000/5,000 units
NsiI-HF	R3127S/L	1,000/5,000 units
PstI-HF	R3140S/L/T/M	10,000/50,000 units
PvuI-HF	R3150S/L	500/2,500 units
PvuII-HF	R3151S/L/M	5,000/25,000 units
SacI-HF	R3156S/L/M	2,000/10,000 units
SalI-HF	R3138S/L/T/M	2,000/10,000 units
SbfI-HF	R3642S/L	500/2,500 units
ScaI-HF	R3122S/L/M	1,000/5,000 units
SphI-HF	R3182S/L/M	500/2,500 units
SspI-HF	R3132S/L/M	1,000/5,000 units
StyI-HF	R3500S/L	3,000/15,000 units
OTHER POPULAR RESTRICTION ENZYMES		
AscI	R0558S/L	500/2,500 units
AvrII	R0174S/L	100/500 units
BglII	R0144S/L/M	2,000/10,000 units
BsaI	R0535S/L	1,000/5,000 units
BsmBI	R0580S/L	200/1,000 units
DpnI	R0176S/L	1,000/5,000 units
MluI	R0198S/L	1,000/5,000 units

Products for Restriction Digestion (Cont.)

PRODUCT	NEB #	SIZE
OTHER POPULAR RESTRICTION ENZYMES (CONT'D)		
NcoI	R0193S/L/T/M	1,000/5,000 units
NdeII	R0111S/L	4,000/20,000 units
NheI	R0131S/L/M	1,000/5,000 units
PacI	R0547S/L	250/1,250 units
PmeI	R0560S/L	500/2,500 units
SmaI	R0141S/L	2,000/10,000 units
SpeI	R0133S/L/M	500/2,500 units
XhoI	R0146S/L/M	5,000/25,000 units
XbaI	R0145S/L/T/M	3,000/15,000 units
XmaI	R0180S/L/M	500/2,500 units
FEATURED GEL LOADING DYE		
Gel Loading Dye, Purple (6X)	B7024S	4 ml
Gel Loading Dye, Purple (6X), No SDS	B7025S	4 ml

For the full list of restriction enzymes available, visit **www.neb.com**.

Products for End Modification

PRODUCT	NEB #	SIZE
Quick Dephosphorylation Kit	M0508S/L	100/500 reactions
Shrimp Alkaline Phosphatase (Recombinant)	M0371S/L	500/2,500 units
Antarctic Phosphatase	M0289S/L	1,000/5,000 units
Alkaline Phosphatase, Calf Intestinal (CIP)	M0290S/L	1,000/5,000 units
T4 DNA Polymerase	M0203S/L	150/750 units
DNA Polymerase I, Large (Klenow) Fragment	M0210S/L/M	200/1,000/1,000 units
Quick Blunting Kit	E1201S/L	20/100 reactions
Mung Bean Nuclease	M0250S/L	1,000/5,000 units
T4 Polynucleotide Kinase	M0201S/L	500/2,500 units
Klenow Fragment (3´ → 5´ exo⁻)	M0212S/L/M	200/1,000/1,000 units

Products for Ligation

PRODUCT	NEB #	SIZE
Blunt/TA Ligase Master Mix	M0367S/L	50/250 reactions
Instant Sticky-End Ligase Master Mix	M0370S/L	50/250 reactions
ElectroLigase	M0369S	50 reactions
T4 DNA Ligase	M0202S/L/T/M	20,000/100,000 units
Taq DNA Ligase	M0208S/L	2,000/10,000 units
Quick Ligation Kit	M2200S/L	30/150 reactions
T3 DNA Ligase	M0317S/L	100,000/750,000 units
T7 DNA Ligase	M0318S/L	100,000/750,000 units

Products for Transformation

PRODUCT	NEB #	SIZE
dam-/dcm- Competent *E. coli*	C2925H/I	20 x 0.05 ml/tube/ 6 x 0.2 ml/tube
NEB 5-alpha Competent *E. coli* (High Efficiency)	C2987H/I/P/R/U	20 x 0.05 ml/tube/ 6 x 0.2 ml/tube/ 1 x 96 well plate 1 x 384 well plate 12 x 8 strip tubes
NEB 5-alpha Competent *E. coli* (Subcloning Efficiency)	C2988J	6 x 0.4 ml/tube
NEB 5-alpha Electrocompetent *E. coli*	C2989K	6 x 0.1 ml/tube
NEB 5-alpha F 1ᵠ Competent *E. coli* (High Efficiency)	C2992H/I	20 x 0.05/6 x 0.2 ml
NEB 10-beta Competent *E. coli* (High Efficiency)	C3019H/I	20 x 0.05 ml/tube/ 6 x 0.2 ml/tube
NEB 10-beta Electrocompetent *E. coli*	C3020K	6 x 0.1 ml/tube
NEB Turbo Competent *E. coli* (High Efficiency)	C2984H/I	20 x 0.05 ml/tube/ 6 x 0.2 ml/tube
NEB Turbo Electrocompetent *E. coli*	C2986K	6 x 0.1 ml/tube

Products for Transformation (Cont.)

PRODUCT	NEB #	SIZE
NEB Stable Competent *E. coli*	C3040S	20 x 0.5 ml/tube

For the full list of competent cells available, visit **www.neb.com**.

Products for Nucleic Acid Purification

PRODUCT	NEB #	SIZE
Monarch Plasmid Miniprep Kit	T1010S/L	50/250 preps
Monarch DNA Gel Extraction Kit	T1020S/L	50/250 preps
Monarch PCR & DNA Cleanup Kit (5 µg)	T1030S/L	50/250 preps

For the list of components available separately, visit **NEBMonarch.com**.

Products for DNA Analysis

PRODUCT	NEB #	SIZE
Quick-Load Purple 1 kb DNA Ladder	N0552S	125 gel lanes
Quick-Load 1 kb DNA Ladder	N0468S/L	125/375 gel lanes
1 kb DNA Ladder	N3232S/L	200/1,000 gel lanes
TriDye 1 kb DNA Ladder	N3272S	125 gel lanes
Quick-Load Purple 100 bp DNA Ladder	N0551S	125 gel lanes
Quick-Load 100 bp DNA Ladder	N0467S/L	125/375 gel lanes
100 bp DNA Ladder	N3231S/L	100/500 gel lanes
TriDye 100 bp DNA Ladder	N3271S	125 gel lanes
Quick-Load Purple 2-Log DNA Ladder	N0550S/L	125-250 gel lanes
Quick-Load 2-Log DNA Ladder	N0469S	250/750 gel lanes
2-Log DNA Ladder (0.1 - 10.0 kb)	N3200S/L	200/1,000 gel lanes
TriDye 2-Log DNA Ladder	N3270S	250 gel lanes
Quick-Load Purple 50 bp DNA Ladder	N0556S	250 gel lanes
Quick-Load 50 bp DNA Ladder	N0473S	125 gel lanes
50 bp DNA Ladder	N3236S/L	200/1,000 gel lanes
Quick-Load 1 kb Extend DNA Ladder	N3239S	125 gel lanes
Quick-Load Purple Low Molecular Weight DNA Ladder	N0557S	125 gel lanes
Quick-Load Low Molecular Weight DNA Ladder	N0474S	125 gel lanes
Low Molecular Weight DNA Ladder	N3233S/L	100/500 gel lanes
Fast DNA Ladder	N3238S	200 gel lanes
PCR Marker	N3234S/L	100/500 gel lanes
Quick-Load PCR Marker	N0475S	125 gel lanes

Products for Seamless Cloning

PRODUCT	NEB #	SIZE
NEBuilder HiFi DNA Assembly Cloning Kit	E5520S	10 reactions
NEBuilder HiFi DNA Assembly Master Mix	E2621S/L	10/50 reactions
Gibson Assembly Cloning Kit	E5510S	10 reactions
Gibson Assembly Master Mix	E2611S/L	10/50 reactions
NEB Golden Gate Assembly Mix	E1600S	15 reactions
BioBrick® Assembly Kit	E0546S	50 reactions
BbsI	R0539S/L	300/1,500 units
BbsI-HF	R3539S/L	300/1,500 units
BsaI	R0535S/L	1,000/5,000 units
BsaI-HF	R3535	1,000/5,000 units
BsmBI	R0580S/L	200/1,000 units
T4 DNA Polymerase	M0203S/L	150/750 units
T5 Exonuclease	M0363S/L	1,000/5,000 units
Exonuclease V (RecBCD)	M0345S/L	1,000/5,000 units
USER™ Enzyme	M5505S/L	50/250 units

Products for Recombinational Cloning

PRODUCT	NEB #	SIZE
Cre Recombinase	M0298S/L/M	50/250 units

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The Next Generation of DNA Assembly and Cloning

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