# Confidently conquer cloning of long DNA fragments

Introducing the TOPO XL-2 Complete PCR Cloning Kit

The Invitrogen<sup>™</sup> TOPO<sup>™</sup> XL-2 Complete PCR Cloning Kit provides all the necessary elements for efficient cloning of extra-long PCR products (up to 13 kb). The kit uses the linearized and topoisomerase I-activated pCR™-XL-2-TOPO™ vector, which is compatible with the cloning of blunt-end PCR fragments (Figure 1). Amplification of long PCR fragments is enabled by Invitrogen™ Platinum™ SuperFi™ Green PCR Master Mix, which is included in the kit. Topoisomerase I activation of the vector enables PCR products to be ligated in just 5 minutes on your benchtop, resulting in high-cloning efficiency (up to 90% positive recombinants).

#### The complete kit includes:

- TOPO XL-2 PCR Cloning Kits containing the pCR-XL-2-TOPO vector
- Platinum SuperFi Green
   PCR Master Mix—featuring a
   proofreading DNA polymerase
   (>100x fidelity compared to Taq
   polymerase) with high processivity
   and a density gradient to generate
   accurate, long PCR amplicons ready
   to load onto an agarose gel for
   gel extraction

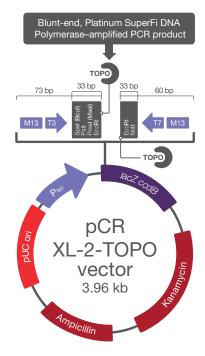
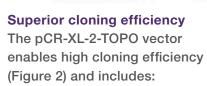
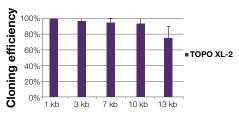


Figure 1. The pCR-XL-2-TOPO vector.

- Invitrogen™ PureLink™ Quick Gel Extraction and PCR Purification Combo Kit—designed to purify DNA fragments in less than 30 minutes from agarose gels or direct PCR purification using a silica-based spin cartridge
- Invitrogen™ One Shot™
   OmniMAX™ 2 T1R Chemically
   Competent E. coli Cells—an
   improved, high-efficiency chemically
   competent cell line, perfect for use
   in all cloning applications



- ccdB gene for positive selection
- EcoRI site flanking the PCR product insertion site for easy excision of inserts
- Ampicillin- and kanamycinresistance genes for your choice of antibiotic selection
- T7 promoter/priming site for in vitro transcription
- T7, T3, and M13 forward- and reverse-primer sites for sequencing



**DNA fragment size** 

Figure 2. TOPO XL-2 PCR cloning kits show high cloning efficiencies. The TOPO XL-2 kit offers superior cloning efficiency for a broad range of different sized targets. Thermo Scientific™ Lambda genomic DNA targets ranging from 1 to 13 kb were cloned using the TOPO XL-2 cloning kit workflow.



## invitrogen

#### Extreme accuracy of your clone

Platinum SuperFi Green PCR Master Mix contains a proofreading DNA polymerase, which combines superior fidelity with the trusted Platinum hot-start technology, designed for the highest success in PCR (Figure 3). Featuring >100x *Taq* fidelity, Platinum SuperFi Green PCR Master Mix is ideally suited for cloning, mutagenesis, and other applications benefiting from supreme sequence accuracy, such as working with long DNA fragments (Figure 4).

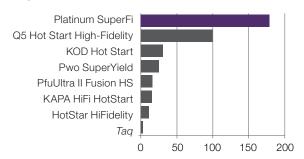


Figure 3. Platinum SuperFi DNA Polymerase exhibits superior fidelity. Relative fidelity values of different DNA polymerases. Polymerase fidelity was measured by next-generation sequencing. The background level of experimental errors was estimated from PCR-free library sequencing data. The polymerase fidelities were normalized to *Taq* polymerase.

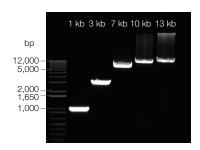


Figure 4. Platinum SuperFi DNA Polymerase has high specificity with amplifying larger targets. 1, 3, 7, 10, and 13 kb targets from Lambda genomic DNA were amplified following manufacturer's standards for PCR cycling parameters.

### **High transformation efficiency**

The OmniMAX 2 T1<sup>R</sup> cells offer the highest transformation efficiency (>5 x  $10^9$  transformants/µg pUC19) of any chemically competent *E. coli* cells in the One Shot format. These highly versatile cells also provide efficient transformation of highly methylated DNA, since OmniMAX 2 T1<sup>R</sup> cells lack the *E. coli* K12 restriction systems (mcrA  $\Delta(mrr\ hsd$ RMS-mcrBC)). In addition, the strain carries the tonA genotype, which confers resistance to T1 and T5 phage infection. This protects your samples and minimizes the possibility of downtime in your lab due to phage contamination (Figure 5).

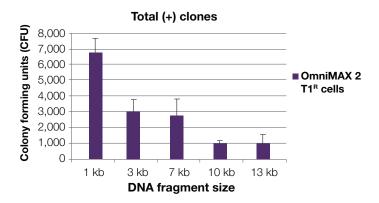


Figure 5. The chemically competent OmniMAX 2 T1<sup>R</sup> cells yield high numbers of CFU. Clones were plated on selection of kanamycin and 1 mM IPTG. 10–12 transformants were analyzed by colony PCR.

Request the TOPO XL-2 Complete PCR Cloning Kit for your Supply Center at **thermofisher.com/scproductrequests** 

#### Ordering information

Product	Size	Cat. No.
TOPO XL-2 Complete PCR Cloning Kit, with One Shot OmniMAX 2 T1R	20 reactions	K8050-20
Chemically Competent E. coli Cells	10 reactions	K8050-10



