



# Efficiently Clone a PCR Product into the Vector of *Your Choice* Using the End-It™ DNA End-Repair Kit

PCR amplification products, produced using Taq Polymerase and other thermostable DNA Polymerases, frequently contain a non-template-coded 3'-A overhang. The End-It™ DNA End-Repair Kit rapidly and efficiently converts PCR product into 5'-phosphorylated, blunt-ended DNA for efficient and economical cloning into any blunt-end cloning vector (Figure 1). The conversion to 5'-phosphorylated, blunt-end DNA is accomplished by exploiting the 5'-3' polymerase and 3'-5' exonuclease activities of T4 DNA Polymerase and T4 Polynucleotide Kinase contained in the End-It End-Repair Enzyme Mix. ATP, dNTPs and 10X Reaction Buffer are also included in the kit. The resulting 5'-phosphorylated, blunt-end DNA can be used directly without purification in a DNA ligation reaction.

The End-It DNA End-Repair Kit is also useful for repairing genomic DNA fragments containing damaged or incompatible 5'- or 3'-ends that result from shearing or restriction endonuclease digestion. The resulting 5'-phosphorylated, blunt-end DNA can be cloned into any blunt-end plasmid, cosmid, fosmid or BAC vector.

**For PCR product cloning, the End-It DNA End-Repair Kit provides:**

### Efficiency

Cloning efficiencies of  $> 10^7$  cfu/ $\mu$ g of PCR product can be obtained (Table 1).

### Versatility

Other PCR cloning methods require cloning into a limited number and type of specialized and costly vectors. Using the End-It DNA End-Repair Kit you can clone your PCR product into the blunt-ended, dephosphorylated vector of your choosing.

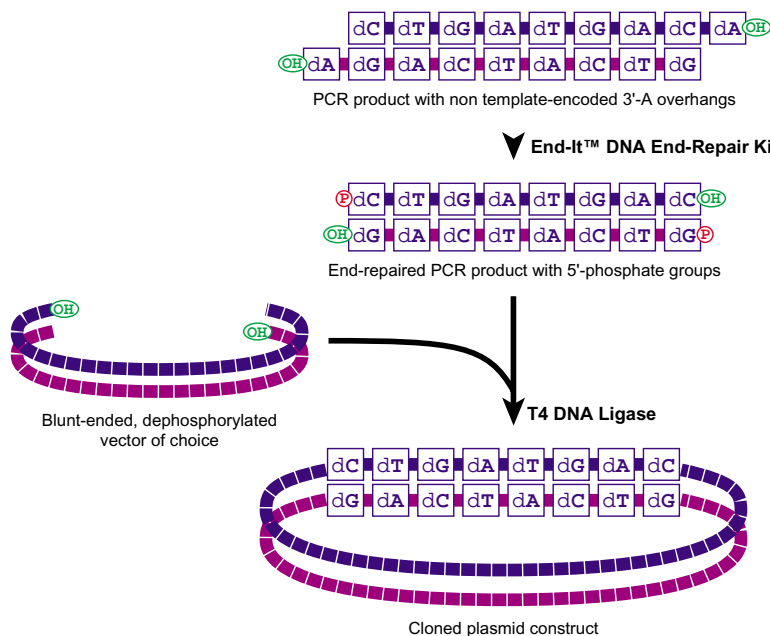
### Economy

One kit will end-repair and 5'-phosphorylate up to 20 PCR products for subsequent blunt-end cloning.

	Colonies/ $\mu$ g DNA
PCR product without treatment	0
PCR product made blunt-ended with End-It Kit	$4 \times 10^7$
Blunt-ended DNA control ( <i>Pvu</i> II-cut DNA)	$4 \times 10^7$

**Table 1. Cloning efficiency of PCR product treated with the End-It™ DNA End-Repair Kit.** 50ng of a 1.3 Kb PCR product, produced using *Taq* DNA polymerase, was ligated into a linearized and dephosphorylated cloning vector before and after end-repair and 5'-phosphorylation using the End-It DNA End-Repair Kit. The positive control was a 1.3 Kb DNA digested with *Pvu* II. Aliquots of the ligation reactions were used to transform TransforMax™ EC100™ Electrocompetent *E. coli*.

**Use the Fast-Link™ DNA Ligation and Screening Kit for rapid and efficient 15 minute blunt-end ligation and 1-hour screening of recombinants. See p. 13.**



**Figure 1. The End-It™ DNA End-Repair Kit converts PCR product with 3'-A overhangs to blunt-end, 5'-phosphorylated DNA for cloning into a blunt-end site of any vector.**

### End-It™ DNA End-Repair Kit

ER0720 20 Reactions

For end-repair and 5'-phosphorylation of up to 100  $\mu$ g of DNA

#### Contents

End-Repair Enzyme Mix, 10X End-Repair Buffer, dNTP Solution, ATP Solution

### TransforMax™ EC100™ Electrocompetent *E. coli*

Highest efficiency electrocompetent cells available. See center insert for more information.

EC10005 5 X 100  $\mu$ l (10 electroporations)

EC10010 10 X 100  $\mu$ l (20 electroporations)