

## Cloning Ready BAC Vectors

### pIndigoBAC-5 (*Bam*H I-Cloning Ready) & pIndigoBAC-5 (*Hind* III-Cloning Ready)

pIndigoBAC-5\* is the first commercially available Bacterial Artificial Chromosome (BAC) vector for cloning and preparation of primary BAC libraries. The vector is derived from pBeloBAC11 and pIndigoBAC<sup>1</sup> and will accommodate and stably maintain DNA inserts of >100 Kb. pIndigoBAC-5 has been linearized at either its unique *Bam*H I or its unique *Hind* III site. The linearized DNA was then dephosphorylated and is ready for cloning the respective *Bam*H I or *Hind* III restriction cut genomic DNA. The linearized and dephosphorylated vectors are highly purified and tested for low vector background and dephosphorylation.

BAC libraries can be produced using the high efficiency TransforMax<sup>TM</sup> EC100<sup>TM</sup> Electrocompetent *E. coli* (available separately; see p. 13). In addition, BAC clones produced in pIndigoBAC-5 can be sequenced directly, without sub-cloning, using the Forward and Reverse BAC end-sequencing primers<sup>2</sup> (available separately) and the EZ::TN<KAN-2> Insertion Kit (see p. 1 for details).

#### Features of pIndigoBAC-5 Cloning Ready vectors

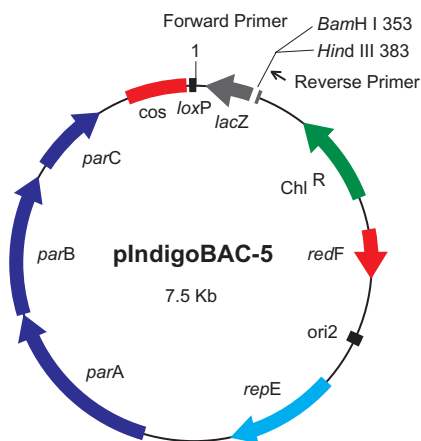
- Derived from pBeloBAC11 and pIndigoBAC.
- Already linearized and dephosphorylated and ready for cloning.
- Tested for low background and for complete dephosphorylation.
- Enhanced blue/white screening of recombinants.

#### Quality control

Greater than 10<sup>6</sup> white colonies/μg of pIndigoBAC-5 DNA are observed when pIndigoBAC-5 (*Bam*H I-Cloning Ready or *Hind* III - Cloning Ready) DNA is ligated with control DNA (*Bam*H I-cut or *Hind* III-cut *E. coli* chromosomal DNA) and transformed into TransforMax<sup>TM</sup> EC100<sup>TM</sup> Electrocompetent *E. coli*. Greater than 95% of white colonies contain an insert and less than 10% background (colonies due to non-linearized pIndigoBAC-5 DNA) is observed.

#### References

1. Birren, B., Mancino, V. and Shizuya, H., (1999) Bacterial Artificial Chromosomes in *Genome Analysis: A Laboratory Manual* v.3, 241-295, Cold Spring Harbor Press.
2. Hurowitz, E.H. et al., (2000) *DNA Research* 7:2,1



**Figure 1. Cloning-Ready pIndigoBAC-5 Vectors are supplied linearized at either the unique *Bam*H I or *Hind* III site and completely dephosphorylated.**

<b>pIndigoBAC-5 (<i>Bam</i>H I-Cloning Ready)</b>		
BACB085H	500ng	25 ng/μl
Supplied linearized at <i>Bam</i> HI site and dephosphorylated.		
<b>pIndigoBAC-5 (<i>Hind</i> III-Cloning Ready)</b>		
BACH095H	500 ng	25 ng/μl
Supplied linearized at <i>Hind</i> III site and dephosphorylated.		
<b>pIndigoBAC-5 Forward Sequencing Primer</b>		
BFP0701	1 nmole	50 μM
<b>pIndigoBAC-5 Reverse Sequencing Primer</b>		
BRP0801	1 nmole	50 μM
<b>TransforMax<sup>TM</sup> EC100<sup>TM</sup> Electrocompetent <i>E. coli</i></b>		
EC10005	5 X 100 μl	(10 Electroporations)
EC10010	10 X 100 μl	(20 Electroporations)
Each includes pUC19 control DNA.		
See p. 13 for information about TransforMax cells.		

\*pIndigoBAC-5 is exclusively licensed by EPICENTRE Technologies.