YEp24

7,769 base pairs
GenBank Accession #: L09156
Not currently available from NEB.

Feature	Coordinates	Source
2µ circle DNA	1-2247	2µ circle plasmid
URA3	2499-3302	*S. cerevisiae*
tet (Tc^R)	3495-4685	pSC101
rop	5326-5517	pMB1
origin (*E. coli*)	6533-5945	pMB1
bla (Ap^R)	7564-6704	*Tn3*

ori = origin of replication
Ap = ampicillin, Tc = tetracycline

There are no restriction sites for the following enzymes: AarI(x), Acc65I, AflII, AgeI, AleI, AscI, AsiSI, AvrII, BaeI, BbvCI, BglII, BsaXI, BsiWI, BsrGI, BssHII, BstEII, BstXI, Bsu36I, FseI, I-CeuI, I-SceI, KpnI, MluI, NotI, PI-PspI, PI-SceI, PacI, PaeR7I, PmeI, PmlI, PspXI, RsrII, SacI, SacII, SanDI(x), SexAI, SfiI, SrfI(x), SwaI, TliI, XhoI.

(x) = enzyme not available from NEB

Yeast **E**pisomal **p**lasmid 24 is a shuttle vector used for gene overexpression in *Saccharomyces cerevisiae*, but also capable of replication in *E. coli*.

While in *E. coli*, the plasmid replicates from the pMB1 origin of replication from pBR322 and is maintained at a similar copy number to pBR322. It carries the *bla* (Ap^R) marker for selection with ampicillin; the tetracycline resistance marker (Tc^R) from pBR322 is also present but is separated from its promoter by the *URA3* sequence, so its expression is variable among different constructions.

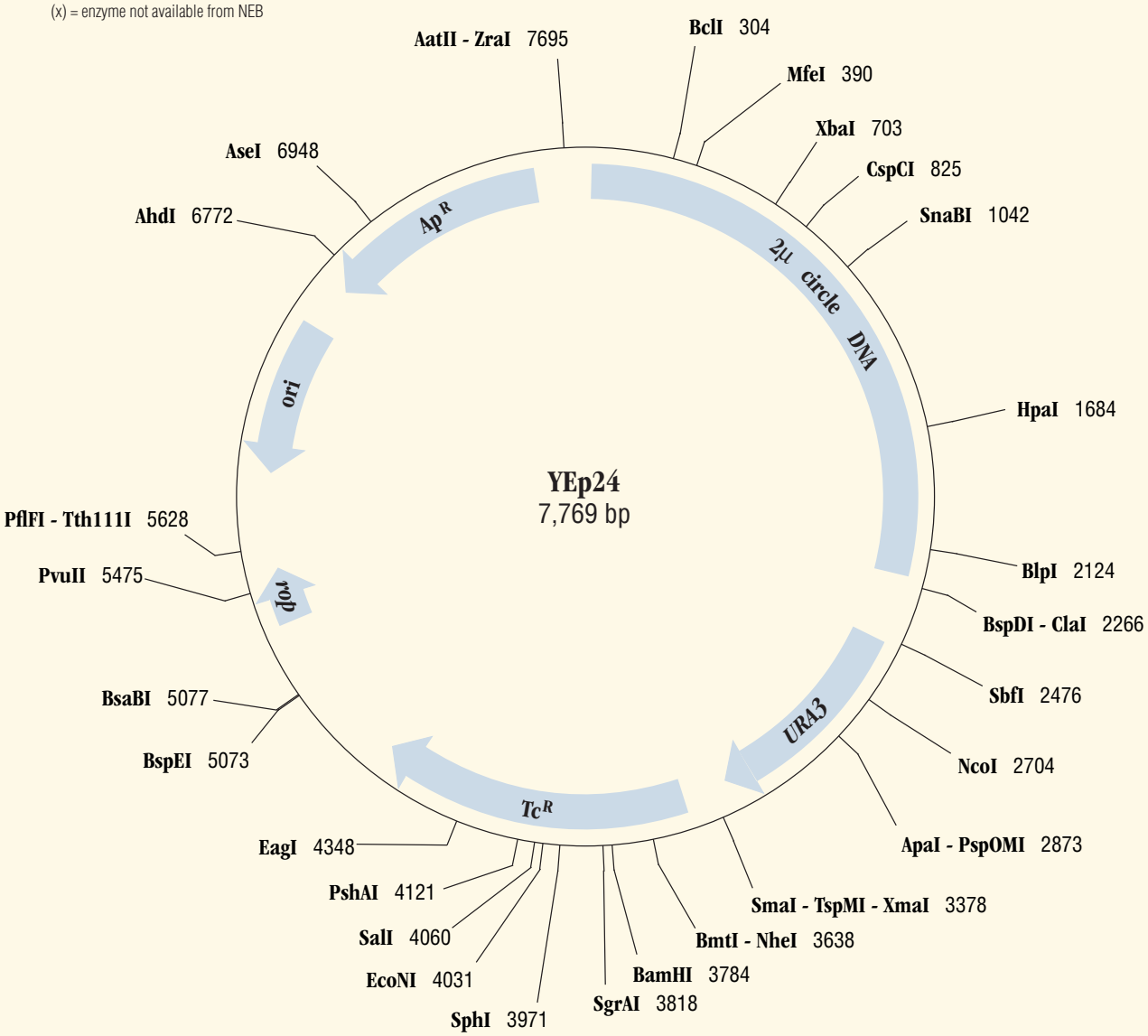
In *S. cerevisiae*, YEp24 replicates at high copy number from the replication determinant of the yeast 2µ circle plasmid (the 2µ circle DNA as marked below is not entirely contiguous within the original 2µ circle plasmid; see GenBank L09156 annotations for details). It carries the *URA3* auxotrophic marker for selection in yeast.

The sequence of YEp24 has been deduced from the sequences of pBR322, the yeast 2µ circle plasmid, and the *URA3* gene with the assistance of R.W. Davis, J. Broach, and D. Botstein (1-5). All sites used in the construction of YEp24 are reconstructed precisely, so the derived sequence should be correct in detail. Numbering of the nucleotide sequence begins at the G in the EcoRI site (...GAATTC...) at the junction between the pBR322 and 2µ circle fragments.

Enzymes with unique restriction sites are shown in **bold** type. Location of sites of all NEB restriction enzymes can be found on the NEB web site (choose Technical Reference > DNA Sequences and Maps). Restriction site coordinates refer to the position of the 5´-most base on the top strand in each recognition sequence.

Open reading frame (ORF) coordinates are in the form "translational start – translational stop"; numbers refer to positions on the top (clockwise) strand, regardless of the direction of transcription and include the start and stop codons.

pMB1 (*E. coli*) origin of replication coordinates include the region from the -35 promoter sequence of the RNAII transcript to the RNA/DNA switch point. *bla* (Ap^R) gene coordinates include the signal sequence.



References

1. Botstein, D. et al. (1979) *Gene* 8, 17–24.
2. Rose, M., Grisafi, P. and Botstein, D. (1984) *Gene* 29, 113–124.
3. Hartley, J.L. and Donelson, J.E. (1980) *Nature* 286, 860–865.
4. Sutcliffe, J.G. (1978) *Cold Spring Harb. Symp. Quant. Biol.* 43, 77–90.
5. Peden, K.W.C. (1983) *Gene* 22, 277–280.