

M13KE

Sequence file available at www.neb.com.
See page 243 for ordering information.

There are no restriction sites for the following enzymes: AarI(x), AatII, AccI, AcuI, AfIII, Agel, AhdI, ApaI, ApaLI, AscI, AsiSI, AvrII, BamHI, BbsI, BcgI, BciVI, BclI, Bclp, BmgBI, BmiI, BsaI, BsgI, BsiWI, BspEI, BspQI, BssHII, BssSI, BstAPI, BstBI, BstEII, BstXI, BstZ17I, Eco53KI, EcoNI, EcoRV, FseI, FspAI(x), HincII, HpaI, I-CeuI, I-SceI, MfeI, MluI, NcoI, NheI, NotI, NruI, NsiI, P1-PspI, P1-SceI, PaeR7I, PflFI, PfiMI, PmeI, PmlI, PshAI, PspOMI, PspXI, RsrII, SacI, SacII, Sall, SanDI(x), SapI, Scal, SexAI, SfiI, SgrAI, SmaI, SpeI, SrfII(x), StuI, StyI, TliI, TspMI, Tth111I, XbaI, XcmI, XhoI, XmaI, ZraI

(x) = enzyme not available from NEB

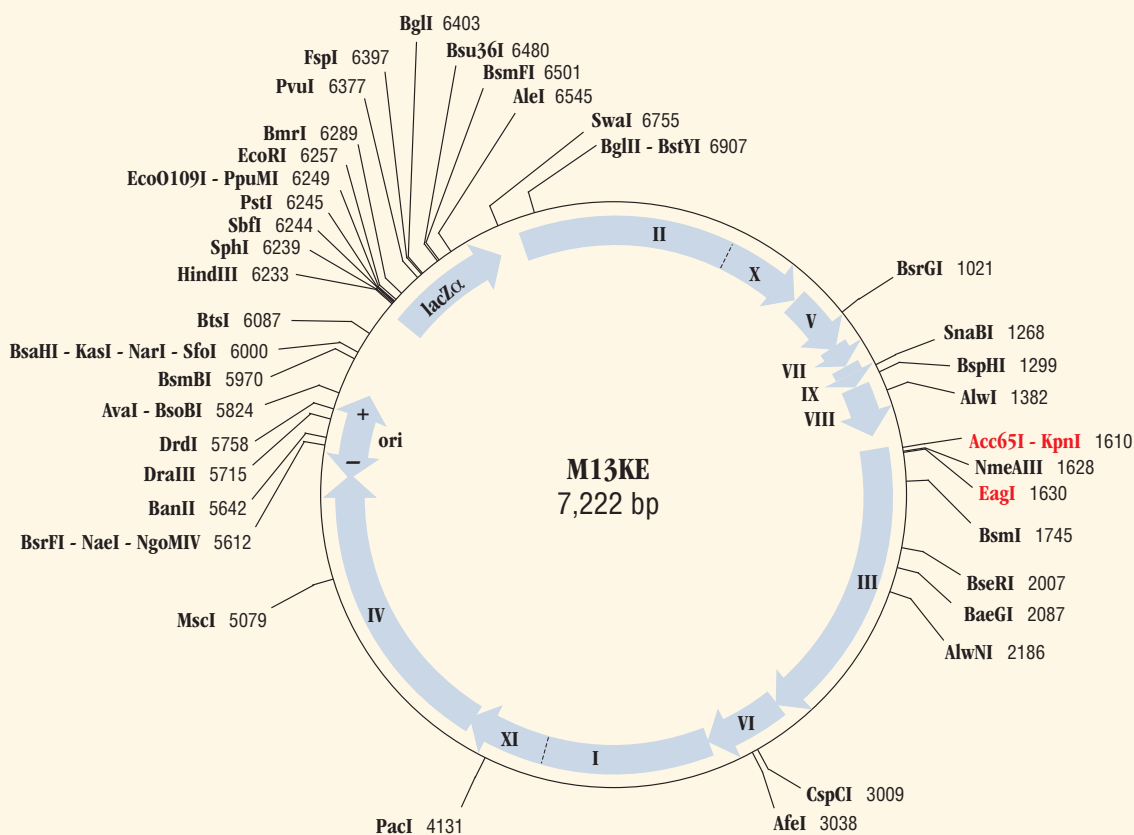
M13KE is a derivative of M13mp19 designed for expression of peptides as N-terminal pIII fusions in phage display applications (1).

Libraries constructed in M13KE are pentavalent (i.e., all five copies of pIII in the mature virion carry the fused peptide). Relative to the parent M13mp19, Acc65I/KpnI and EagI sites (red) have been introduced flanking the pIII leader peptidase cleavage site, and the Acc65I/KpnI site in the multiple cloning site (MCS) was deleted. Phage displayed random peptide libraries are constructed by annealing an extension primer to a synthetic oligonucleotide encoding the random peptide library and a portion of the pIII leader sequence, extending with DNA polymerase, and digesting with Acc65I and EagI (2). The resulting cleaved duplex is inserted into M13KE which has been digested with the same enzymes. M13KE and the insert extension primer are supplied with the Ph.D. Peptide Display Cloning System (NEB #E8101).

The complete nucleotide sequence of M13KE has been determined at New England Biolabs, resulting in several nucleotide changes relative to the previous, deduced sequence (3).

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.

M13 origin of replication arrows indicate the direction of synthesis of both the (+) and (-) strands.



References

- (1) Zwick, M.B. et al. (1998) *Anal. Biochem.*, 264, 87–97.
- (2) Noren, K.A. and Noren, C.J. (2001) *Methods*, 23, 169–178.
- (3) Stewart, F.J. (2002) unpublished observations.