



cDNA Library, Rat Embryonic Fibroblast

BACKGROUND

This cDNA library (plasmid DNA) is constructed from rat embryonic fibroblast-derived poly(A)⁺ RNA by the Linker-Primer method (Ref.1) by Professor Hiroshi Nojima of Research Institute for Microbial Diseases, Osaka University. This library is unidirectionally cloned by using the oligo (dT)₁₈ linker primer which contains the restriction enzyme site of *Not* I, and *Bam*HI (*Bgl* II)-*Sma* I adaptor.

The pAP3*neo* vector used in this library can express rat genes in mammalian cells as it contains SV40 promoter. It also contains Ori of pUC plasmid required for replication in *E.coli*, f1 ori which is necessary for ssDNA synthesis, and bacteriophage T7 and T3 promoters for RNA synthesis (see Figure). GenBank Accession No. AB003468

Applications:

PCR screening of known or unknown gene: Prepare the primers for the known or unknown gene (cDNA) and amplify the gene by PCR from this library, followed by cloning to an appropriate vector. It is useful for large-scale protein productions, and preparation of probes, etc.

Standard amplifying conditions: 35 cycles of PCR reactions using 10-100 ng of cDNA as a template. (Change the quantity of template and the number of cycles depending on the expression rate of mRNA of the objective gene.)

Size:

500 ng (40 ng/ul, 13ul) in 10 mM Tris-HCl-1mM EDTA (pH 7.5)

Quality:

- 1) Number of independent clones: 62×10^6
- 2) Average insert size : longer than 1 kb

Storage:

Store at -20°C

References:

- 1) Kobori M *et al* " Large scale isolation of osteoclast-specific genes by an improved method involving the preparation of a subtracted cDNA library." *Genes Cells* **3**: 459-475 (1998) PMID: 9753427
- 2) Sambrook J and Russell DW *Molecular Cloning* Chapter 11 "Preparation of cDNA libraries and gene identification." CSHL Press (2001)

Note

- * This library is to be used only by the purchaser. It is not allowed to amplify and transfer the library to a third person.
- * Related products: human tissue specific cDNA libraries and cDNA libraries of model organisms.
- * For custom order of cDNA cloning from the libraries, construction of protein expression systems, and production and purification of proteins. Contact with export@cosmobio.co.jp

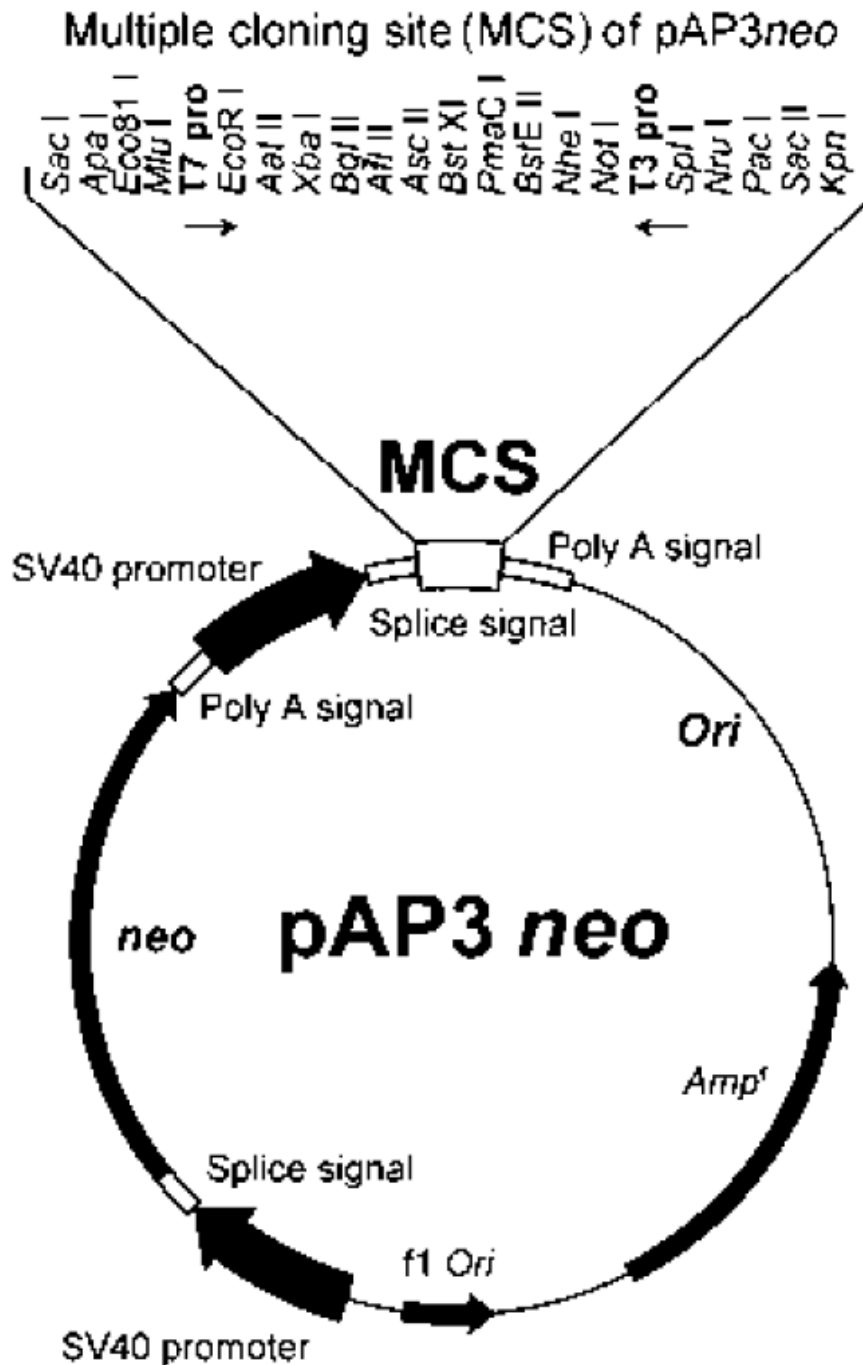


Fig. Structure of pAP3neo and the restriction sites

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