cDNA Library, *S. cerevisiae* , Log Phase

02-701　　500 ng

This cDNA library (plasmid DNA) is constructed from *Saccharomyces cerevisiae*, strain S288C-derived poly(A)$^+$ RNA at the log phase by the Linker-Primer method (Ref.1) by Prof. H. Nojima of 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 pLZ3 vector (shown below) used in this library can not replicate in *S. cerevisiae* but contains pUCori for replication in *E. coli*

Application

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.

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.)

Specification

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

Quality: 1) Number of independent clones: 3.6×10^6

2) Average insert size : longer than 1 kb

Storage: -20℃

References: Construction of this library is described in Supplementary data of Ref.3

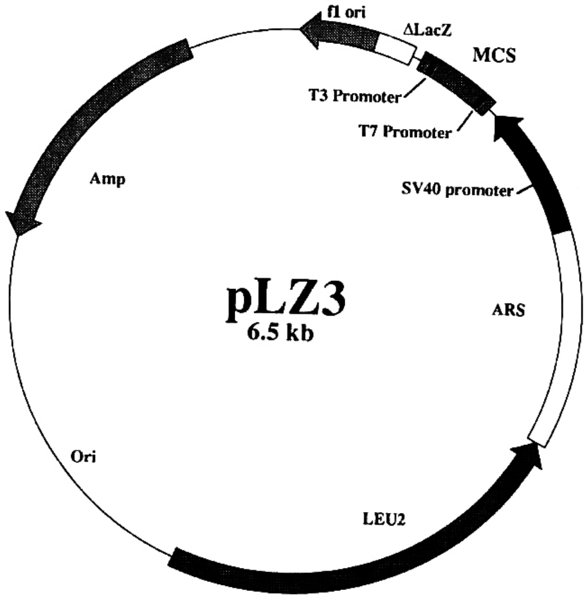
1. Kobori M *et al* "Large scale isolation of osteoclast-specific genes by an improved method involving the preparation of a subtracted cDNA library." *Genes Cells* **3**: 459-475 (1998) PMID: 9753427
2. Tanaka S and Nojima H "Nik1: a Nim1-like protein kinase of *S. cerevisiae* interacts with the Cdc28 complex and regulates cell cycle progression." *Genes Cells* **1**, 905-921 (1996) PMID: 9077450
3. Tougan T, Okuzaki D, Nojima H. Chum-RNA allows preparation of a high-quality cDNA library from a single-cell quantity of mRNA without PCR amplification. Nucleic. Acids Res., 36(15):e92, (2008) PMID:18603591

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 (See HP).

to be continued…

Fig. Structure of pLZ3 and the restriction sites.



; MCS(pLZ3)

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                      CpoI(3)    SauI(b) MluI(5)                              AatII(3) BglII(5)  AscI(5)        BalI(b)
             PstI(3)SacI(3)    ApaI(3)   ------                               ------ ------    --------       ------
              SseI(3)------    ------------        T7  Promoter     EcoRI(5)   XbaI(5)AflII(5)      BstXI(5)
              --------  -------  -------        ------------------ ------     ------ ------     ------------ --
NNNCTGCA  CCTGCAGGAGCTCGGACCGGGCCCTTAGGACGCGTAATACGACTCACTATAGGGAATTCGACGTCTAGATCTTAAGGCGCGCCAAGGGGTTGGCCA
NNNG   ACGTGGACGTCCTCGAGCCTGGCCCGGGAATCCTGCGCATTATGCTGAGTGATATCCCTTAAGCTGCAGATCTAGAATTCCGCGCGGTTCCCCAACCGGT

   BstEII(5)
   -------        NheI(5)                                                   SwaI(3)       NruI(b)       SacII(3)
SnaBI(b)  DraIII(3) ------     SceI(3)            NotI(5)  T3 promoter --------SplI(5)------ PacI(3)------  SacI(3)
 ------   -----------      ------------------         -------------------      ------        --------       ------
 CGTGGTAACCACGGGGTGGCTAGCTAGGGATAACAGGGTAATATAGCGGCCGCCCTTTAGTGAGGGTTAATTTAAATCGTACGTCGCGATTAATTAACCGCGGTGGAGCT  CAAT
 GCACCATTGGTGCCCCACCGATCGATCCCTATTGTCCCATTATATCGCCGGCGGGAAATCACTCCCAATTAAATTTAGCATGCAGCGCTAATTAATTGGCGCCACC  TCGACTTA

 TCGCCCTATAGTGAGTCGTATTA -3'
 AGCGGGATATCACTCAGCATAAT -5'
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