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. cereviseaas* 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 x 10^6

   2) Average insert size : longer than 1 kb

  Storage: -20

References

1. Kobori,M., Ikeda,Y., Nara,H., Kato,M., Kumegawa,M., Nojima,H., and Kawashima,H. " 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. 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 (See HP).

Fig. Structure of pLZ3 and the restriction sites. Ars is the region required

for replication in S. pombe, and **Ori** is a plasmid origin for replication in *E. coli*

f1 ori
ΔLacZ
MCS
T3 Promoter
T7 Promoter
SV40 promoter
Amp
pLZ3
6.5 kb
ARS
Ori
LEU2
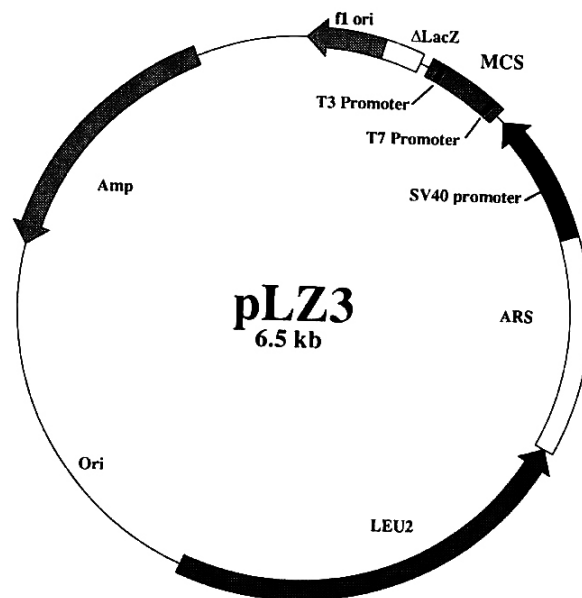
; MCS(pLZ3)

```
              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'
```