

Microbial Genomic DNA

Strain: JCM 20832T
Organism: *Halobacillus halophilus*

RIKEN DNA Bank catalog #

JGD 12238

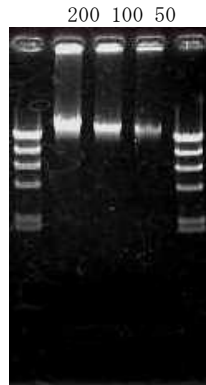
Lot # : 12238-A3I9

| | | |
|---------------------|----|-------|
| Final concentration | 50 | ng/ul |
| Shipping amount | 20 | ul |

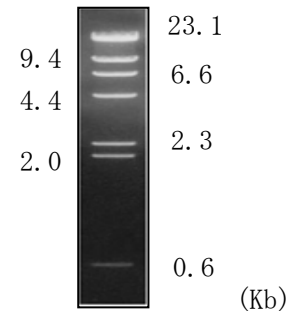
1. Electrophoresis of Genomic DNA

Date : 2013/11/5

Electrophoresis : 1% agarose gel, 1x TAE buffer
Loading DNA amount : 200, 100 and 50 ng/lane
Size marker : Lambda/HindIII (200 ng/lane)



Marker
Lambda/HindIII
(NEB#N3012S)



2. PCR Amplification

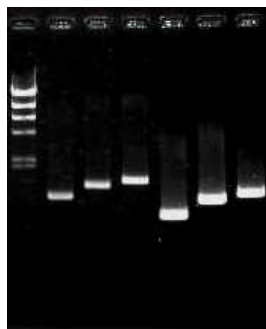
Date : 2013/11/5

Conditions of PCR ribosomal RNA (rRNA) gene

Template : 50 fold diluted DNA, 1 ul
Enzyme : KOD Plus (Toyobo)
Volume : 20 ul

Reaction program: 94 degC 2 min
x30 cycle $\left\{ \begin{array}{l} 94 \text{ degC } 15 \text{ sec} \\ 60 \text{ degC } 30 \text{ sec} \\ 68 \text{ degC } 60 \text{ sec} \end{array} \right.$
68 degC 2 min

(1) (2) (3) (4) (5) (6)



PCR primers

| 16S rRNA Eubacteria set | |
|-------------------------|-------------------------|
| EB-20F | 5' AGTTTGATCCTGGCTC 3' |
| EB-350F | 5' TACGGGAGGCAGCAG 3' |
| EB-1100R | 5' AGGTTGCGCTCGTTG 3' |
| EB-1400R | 5' ACGGGCGGTGTGTAC 3' |
| EB-1530R | 5' AAGGAGGTGATCCAGCC 3' |

Sample Number

| | EB-20F | EB-350F |
|----------|--------|---------|
| EB-1100R | (1) | (4) |
| EB-1400R | (2) | (5) |
| EB-1530R | (3) | (6) |

Electrophoresis : 1% agarose gel, 1x TAE buffer
Loading DNA amount : 5 ul PCR product/lane
Size marker : Lambda/HindIII (200 ng/lane)

3. Nucleotide sequence of rRNA gene

 Confirmed date : 2013/11/7