

Microbial Genomic DNA

Strain : **JCM 7540**
 Organism : ***Paenibacillus thiaminolyticus***

RIKEN DNA Bank catalog #
 JGD : **08471**
 Lot # : **15770_A8Ao**

1. Shipping

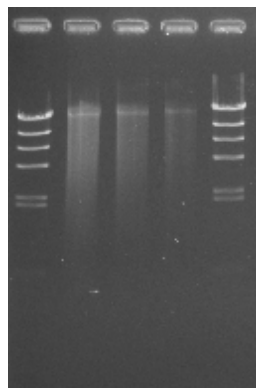
Volume : **20** uL

2. Concentration of Genomic DNA

Concentration (OD 260) : **181** ng/uL
 A260 / A280 : **1.9**

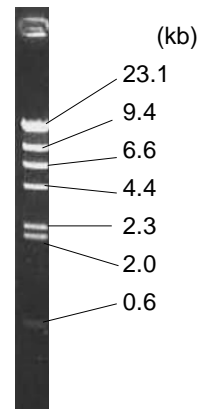
3. Electrophoresis of Genomic DNA

Electrophoresis : 1% agarose gel, 1x TAE buffer
 Loading DNA amount : **2.0, 1.0, 0.5** uL/lane
 size marker : Lambda / HindIII (200 ng/Lane)
2.0, 1.0, 0.5 (uL)



Date : **2018/2/8**

Marker (Lambda / HindIII, NEB#3012S)

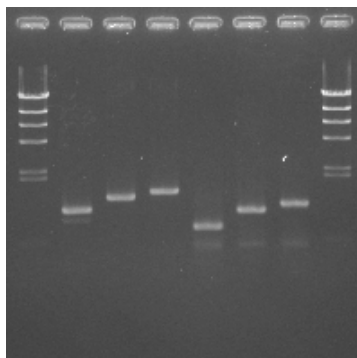


4. PCR Amplification

Conditions of PCR Ribosomal RNA (rRNA) gene
 Template : 100 fold diluted DNA, 1 uL
 Enzyme : KOD plus (TOYOBO)
 Volume : 20 uL

Reaction program :
 94 degC 2 min
 x 30 cycle $\left[\begin{array}{ll} 94 \text{ degC} & 15 \text{ sec} \\ 60 \text{ degC} & 30 \text{ sec} \\ 68 \text{ degC} & 60 \text{ sec} \end{array} \right]$
 68 degC 2 min

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



Date : **2018/2/8**

PCR primers for rRNA gene

| | |
|----------|---------------------------|
| EB-20F | 5'- AGTTTGATCCTGGCTC -3' |
| EB-350F | 5'- TACGGGAGGCAGCAG -3' |
| EB-1100R | 5'- AGGGTTGCGCTCGTTG -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 : 3 uL PCR product/Lane
 Size marker : Lambda / HindIII (200ng/Lane)

5. Nucleotide sequence of rRNA gene

Confirmed Date : **2018/2/9**