

**Microbial Genomic DNA**

RIKEN DNA Bank catalog #

JGD 08101

Lot # : 110629

Strain: JCM 9141

Organism: *Bacillus pseudofirmus*, A-40-2

Final concentration : 100 ng/ul

Shipping amount : 100 ul

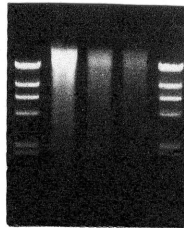
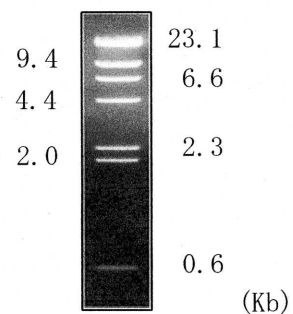
## 1. Electrophoresis of Genomic DNA

Date : 2011/6/28

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 : 2011/6/29

Conditions of PCR (16S ribosomal RNA genes)

Template : 100 fold diluted DNA, 1 ul

Enzyme : KOD Plus (Toyobo)

Volume : 20 ul

Reaction program: 94 degC 2 min  
 x30 cycle ( 94 degC 15 sec )  
 ( 60 degC 30 sec )  
 ( 68 degC 60 sec )  
 ( 68 degC 2 min )

PCR primers

Eubacteria set	
EB-20F	5' AGTTGATCCTGGCTC 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 : 5 ul PCR product/lane

Size marker : Lambda/HindIII (200 ng/lane)

## 3. Nucleotide sequence of 16S rRNA gene

 Confirmed date : 2011/7/7