

# Microbial Genomic DNA

Strain: JCM 13517T  
Organism: *Thermococcus onnurineus*

RIKEN DNA Bank catalog #

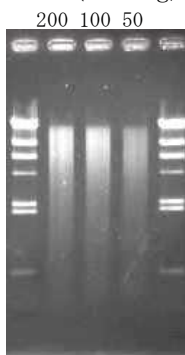
JGD 08242  
Lot # : 110920

Final concentration : 100 ng/ul  
Shipping amount : 20 ul

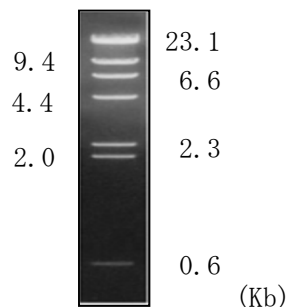
## 1. Electrophoresis of Genomic DNA

Date : 2011/9/21

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)



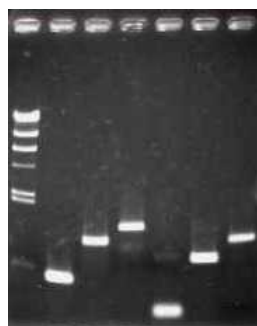
## 2. PCR Amplification

Date : 2011/9/22

Conditions of PCR (16S ribosomal RNA genes)

Template : 1 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)



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

PCR primers

Archea set	
A-20F	5' TCCGGTTGATCCTGCCG 3'
A-340F	5' CCCAGGCCCTACGGG 3'
A-520R	5' GTATTACCGCGGCGGCTG 3'
A-1100R	5' CGGGTCTCGCTCGTT 3'
A-1400R	5' GACGGGCGGTGTGTGC 3'

Sample Number

	A-20F	A-340F
A-520R	(1)	(4)
A-1100R	(2)	(5)
A-1400R	(3)	(6)

## 3. Nucleotide sequence of 16S rRNA gene

Confirmed date : 2011/9/28