

Microbial Genomic DNA

Strain : **JCM 20666**
Organism : ***Alcaligenes faecalis***

RIKEN DNA Bank catalog #
JGD : **07089**
Lot # : **07089_A7K6**

1. Shipping

Volume : **20** μ L

2. Concentration of Genomic DNA

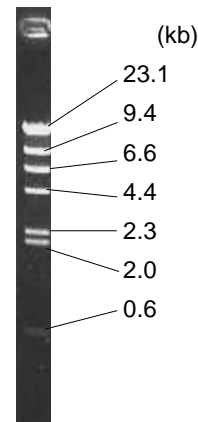
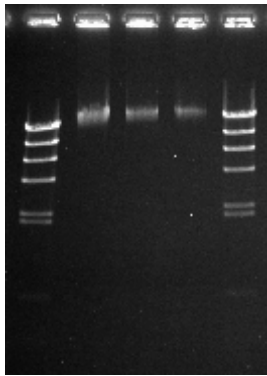
Concentration (OD 260) : **227** ng/ μ L
A260 / A280 : **1.92**

3. Electrophoresis of Genomic DNA

Electrophoresis : 1% agarose gel, 1x TAE buffer
Loading DNA amount : **1,0.5,0.25** μ L/lane
size marker : Lambda / HindIII (200 ng/Lane)
1, 0.5, 0.25 (μ L)

Date : **2017/11/6**

Marker (Lambda / HindIII, NEB#3012S)



4. PCR Amplification

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

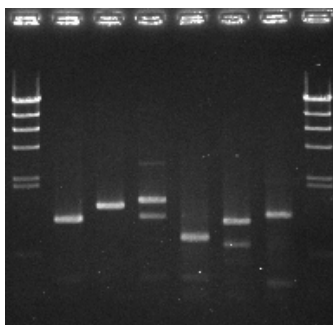
Reaction program :
x 30 cycle $\left[\begin{array}{ll} 94 \text{ degC} & 2 \text{ min} \\ 94 \text{ degC} & 15 \text{ sec} \\ 60 \text{ degC} & 30 \text{ sec} \\ 68 \text{ degC} & 60 \text{ sec} \\ 68 \text{ degC} & 2 \text{ min} \end{array} \right]$

Date : **2017/11/6**

PCR primers for 16S rRNA

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'

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



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 μ L PCR product/Lane
Size marker : Lambda / HindIII (200ng/Lane)

5. Nucleotide sequence of rRNA gene

Confirmed Date : **2017/11/7**