

Microbial Genomic DNA

Strain : **JCM 5491**
Organism : ***Escherichia coli***

RIKEN DNA Bank catalog #

JGD : **12466**Lot # : **12466_B1J6**

1. Shipping

Volume : **100** uL

2. Concentration of Genomic DNA

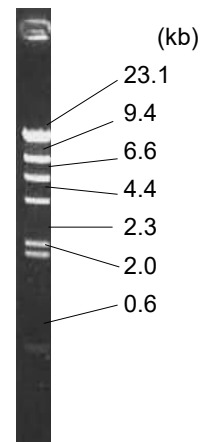
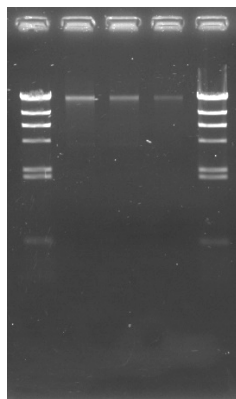
Concentration (OD 260) : **15** ng/uL
A260 / A280 : **1.92**

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 : **2021/10/7**

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 :
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 : **2021/10/7**

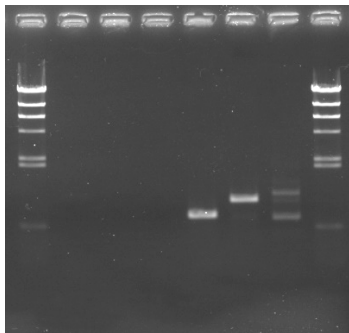
PCR primers for rRNA gene

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'

Reference: Please visit online-manual site below.

http://dna.brc.riken.jp/en/manualen/manual_genom_dna

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

5. Nucleotide sequence of rRNA gene

Confirmed Date : **2021/10/8**