

# Microbial Genomic DNA

Strain : **JCM 5963<sup>T</sup>**  
 Organism : ***Pseudomonas fluorescens***

RIKEN DNA Bank catalog #  
**JGD : 08021**  
**Lot # : 08021\_A6D4**

## 1. Shipping

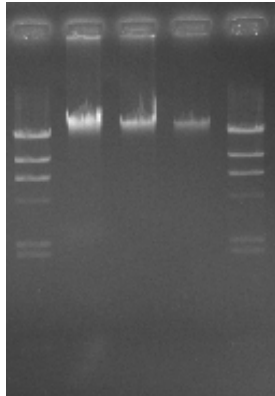
Volume : **20** uL

## 2. Concentration of Genomic DNA

Concentration ( OD 260 ) : **70** ng/uL  
 A260 / A280 : **1.92**

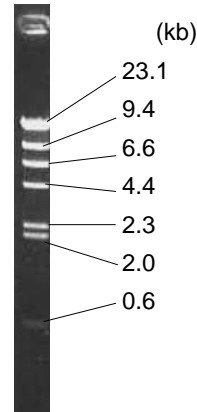
## 3. Electrophoresis of Genomic DNA

Electrophoresis : 1% agarose gel, 1x TAE buffer  
 Loading DNA amount : **4, 2, 1** uL/lane  
 size marker : Lambda / HindIII ( 200 ng/Lane )  
**4, 2, 1** ( uL )



Date : **2016/4/6**

Marker ( Lambda / HindIII, NEB#3012S )

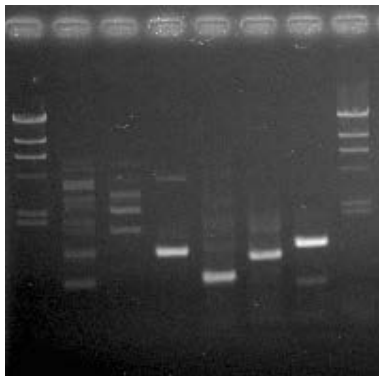


## 4. PCR Amplification

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 : **2016/4/7**

### 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'

### 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 : **2016/4/11**