

# 1 kb DNA Ladder



1-800-632-7799  
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www.neb.com



N3232S 114130515051

## N3232S

200 gel lanes (100 µg) Lot: 1141305 Exp: 5/15  
500 µg/ml Store at -20°C

1.5 ml Gel Loading  
Dye, Blue (6X) Store at 25°C

**Description:** A number of proprietary plasmids are digested to completion with appropriate restriction enzymes to yield 10 bands suitable for use as molecular weight standards for agarose gel electrophoresis. The digested DNA includes fragments ranging from 0.5–10.0 kilobases (kb). The 3.0 kb fragment has increased intensity to serve as a reference band.

Supplied in: 10 mM Tris-HCl (pH 8.0),  
1 mM EDTA.

**Reagents supplied:**  
6X Gel Loading Dye, Blue

**1X Gel Loading Dye, Blue:**  
2.5% Ficoll-400  
11 mM EDTA  
3.3 mM Tris-HCl (pH 8.0@25°C)  
0.017% SDS  
0.015% bromophenol blue

**Preparation:** The double-stranded DNA is digested to completion with appropriate restriction enzymes, phenol extracted and equilibrated to 10 mM Tris-HCl (pH 8.0) and 1 mM EDTA.

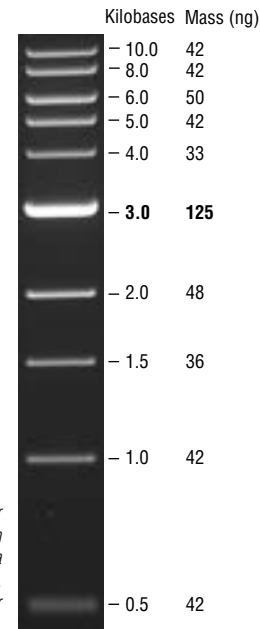
**Usage Recommendation:** The 1 kb DNA Ladder was not designed for precise quantification of DNA mass, but can be used for approximating the mass of DNA in comparably intense samples of similar size. The approximate mass of DNA in each of the bands in our 1 kb DNA Ladder is as follows (assuming a 0.5 µg loading):

Fragment	Base Pairs	DNA Mass
1	10,002	42 ng
2	8,001	42 ng
3	6,001	50 ng
4	5,001	42 ng
5	4,001	33 ng
6	<b>3,001</b>	<b>125 ng</b>
7	2,000	48 ng
8	1,500	36 ng
9	1,000	42 ng
10a	517	42 ng
10b	500	

**Notes:** All fragments have a 4-base, 5' overhangs that can be end labeled using T4 Polynucleotide Kinase (NEB #M0201) or filled-in using DNA Polymerase I, Klenow Fragment (NEB #M0210) (1). Use  $\alpha$ -[<sup>32</sup>P] dATP or  $\alpha$ -[<sup>32</sup>P] dTTP for the fill-in reaction.

1 kb DNA Ladder is stable for at least 3 months at 4°C.

For long term storage, store at -20°C. If samples need to be diluted, use TE or other buffer of minimal ionic strength. DNA may denature if diluted in dH<sub>2</sub>O.



1 kb DNA Ladder  
visualized by ethidium  
bromide staining on a  
0.8% TAE agarose gel.  
Mass values are for  
0.5 µg/lane.

(see other side)

CERTIFICATE OF ANALYSIS

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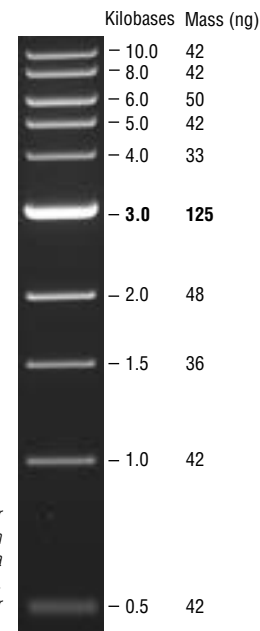
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CERTIFICATE OF ANALYSIS

Due to the limitations of the acrylamide gel technology, one or two extra bands may be visible on the DNA ladders when run on a polyacrylamide gel.

**Suggested protocol for loading a sample:**

The following protocol is recommended for a 5 mm wide lane.

1. Prepare loading mixture:

Distilled water	4 $\mu$ l
6X Blue Loading Dye	1 $\mu$ l
DNA Ladder	1 $\mu$ l
Total volume	6 $\mu$ l

2. Mix gently
3. Load onto the agarose gel

**Note:** The components of the mixture should be scaled up or down, depending on the width of the agarose gel.

**Reference:**

1. Sambrook, J., Fritsch, E. F. and Maniatis, T. (1989). *Molecular Cloning: A Laboratory Manual*, (2nd ed.), (pp. 10.51–10.67). Cold Spring Harbor: Cold Spring Harbor Laboratory Press.

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