

# Lambda *Hind*III Ladder

The  $\lambda$  *Hind*III DNA Ladder is prepared by restriction digestion of phage Lambda DNA to completion with *Hind*III, followed by heat inactivation of the enzyme.

The resulting 7 fragments range from 564 base pairs to 23.13 kilobases in size.

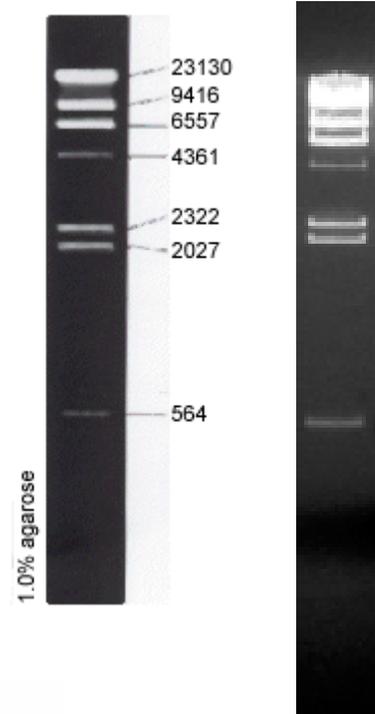
564, 2027, 2322, 4361, 6557, 9416, 23130 bp

(A 125 bp fragment is present but usually not seen.)

The  $\lambda$  *Hind* DNA Ladder is supplied ready to use in loading buffer containing dye.

Load 10 to 15 ml onto the gel (340 to 500 ng)

Recommended concentration of agarose 0.7 to 1.7%.



**1 kb  
DNA Ladder**



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**N3232S**

100  $\mu$ g

Lot: 54

500  $\mu$ g/ml

Store at  $-20^{\circ}\text{C}$

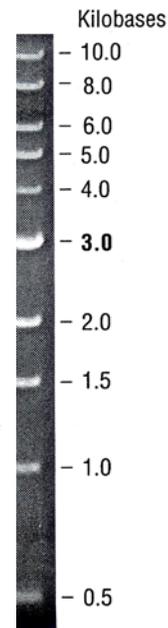
**Description:** A number of proprietary plasmids are digested to completion with appropriate restriction enzymes to yield 10 bands suitable for use as molecular weight standards for agarose gel electrophoresis. The digested DNA includes fragments ranging from 0.5–10.0 kilobases (kb). The 3.0 kb fragment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

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

**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  $\mu$ g 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	



**6X loading dye** contains:

*Bromophenol Blue* (migrates around **300 bp** in 1% agarose)

*Xylene Cyanol* (migrates around **4 kbp** in 1% agarose)

40% sucrose, which gives it density

Antibiotic-containing plates:

Black/Black = kanamycin

Black/Red = ampicillin

Black/Black/Red = kan + amp

1 unit restriction enzyme = activity to digest 1  $\mu\text{g}$  of DNA in 50  $\mu\text{L}$  reaction volume in 1 hour

Transformation efficiency = colonies/ $\mu\text{g}$  DNA transformed

e.g. plasmid DNA solution: 10  $\mu\text{g}/\text{mL}$  = 10  $\text{ng}/\mu\text{L}$

Transform 5  $\mu\text{L}$  = 50  $\text{ng}$

Plate 100  $\mu\text{L}$  (out of 1  $\text{mL}$  total volume) = 10% = 5  $\text{ng}$

Get 50 colonies

Transformation efficiency = 50 colonies / 5  $\text{ng}$  =  $1 \times 10^4$  colonies /  $\mu\text{g}$