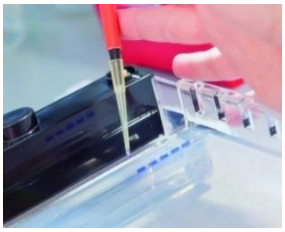
Technical Info

Well advised with Roth.



DNA Size Markers

Tipps for Application

DNA Size Markers and Ladders

Roth offers a multifaceted marker range including DNA markers for each application required.
Our DNA-markers produce sharp, clear bands. Terminally, the unregular markers carry sticky *Eco*R I sites and, therefore, end-labelling methods can be applied easily.

The DNA-markers are supplied ready-to-use or in lyophilised form. Gel loading buffer for dissolving the lyophilised DNA-fragments is included. ROTI®Load DNA with glycerol (Art. No. 0100.1), or the specially designed short-run-loading buffer (Art. No. 0099.1) is used as gel loading buffer).

Shipping: All our DNA size markers are delivered at ambient temperature. The ‚dry' marker versions contain highly stabile DNA. In the solubilized ready-to-use markers, the DNA is stabilized quite well by the used gel loading buffer formulation and may be stored – or shipped – for many days or some weeks at moderate ambient temperature.

Storage: Store marker in small aliquots at -20 °C. One aliquot can be stored at +4 °C for permanent use.

Amount of applied DNA: Apply more marker when markers with higher band numbers are used. Applied amount can be reduced, however, for short separation distances. Application on polyacrylic amide gels may be reduced to 1/5 to ½ of the marker amount used for agarose gels

Fast running gels: Roth markers are established with respect to high separation efficiency even when applying high voltages. Be sure, however, to use sufficient and fresh gel running buffer made with water of good quality.

Dye concentration: IN case the dye bands are too prominent and tend to overlay important DNA fragments, dilute the marker using 1x TE up to 1:2 (ROTI®Stock 100 x TE, Art. No. 1052.1).

For **direct detection of the marker DNA in the gel under UV- or blue light** we recommend to use lyophilized marker DNA. The marker DNA is first solubilized in sterile water or 1xTE buffer, then ROTI®Load DNAstain (6x conc.) is added. The so prepared marker may be aliquoted and stored at -20 °C. ROTI®Load DNAstain contains a fluorecent dye which may be excited by UV- or blue light, emitting at 525 nm.

‘Falsely' running bands: Under extreme conditions, (e.g. high voltages, short running distances, salt content in the probes >150 mM) implausible results may occur, which are often caused by partial strand separation of the DNA. For amendments we recommend: a) Supplementation of the marker with restriction enzyme buffer in order to increase the salt content, b) use lower separation voltages, c) make sure to efficiently remove evolving heat, e.g. by cooling the gel prior to or during the run.

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Tips for Estimating the Amount of DNA in Agarose Gels

- Please make sure to quantitatively dissolve the comparative DNA (e.g. the marker) in buffer, in order to obtain the full DNA-concentration.
- Allow the gel to run far enough so that the bands have adequate space, the DNA can then disperse in the bands and stain well.
- Stain the gel thoroughly - we recommend staining after the gel run. Staining the DNA by adding ethidium bromide to the agarose will not achieve an even distribution in the gel and is frequently not quantitative.
- Apply several lanes of comparative-DNA with different quantities so that various bands can be compared. A calibration curve may also be plotted to determine the exact concentration.
- For comparison it is best to use bands of similar size to your DNA (maximum ±50 % of the fragment length).
- Ethidium bromide is integrated much more effectively between the base pair stacks in linear DNA than in cyclic DNA. Therefore, please make sure that the conformation of the DNA whose concentration you wish to estimate is identical to that of the comparative DNA. In the event of cyclic DNA, i.e. isolated plasmids, cyclic DNA should also be used, e.g. plasmid-DNA pUC19 (Art. No. X911.1) or pBR322 (Art. No. X912.1). If you are using a DNA-marker as a comparative DNA and wish to measure cyclic DNA, you should initially linearize an aliquot of the DNA through restriction digestion and apply the preparation to the gel.
- In the event of a Lambda/*Hin*d III marker: Do not use the 4.4 kb band of the marker for concentration analysis, as it is often not quantitatively present but coupled to the large fragment via the cos-site. Should you require the 4.4 kb band, please take care to heat the marker prior to applying (5 minutes at 65 °C).
- Only use the large marker fragments of a Lambda marker for concentration estimation when your fragments are in the same size range. Many visual systems do not depict the staining of the large bands proportionally to the DNA-amount.

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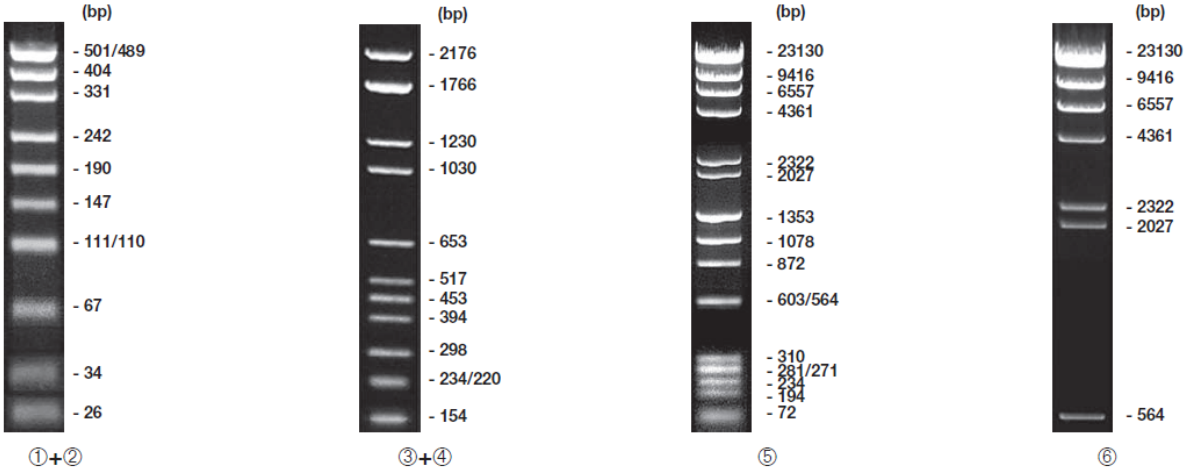
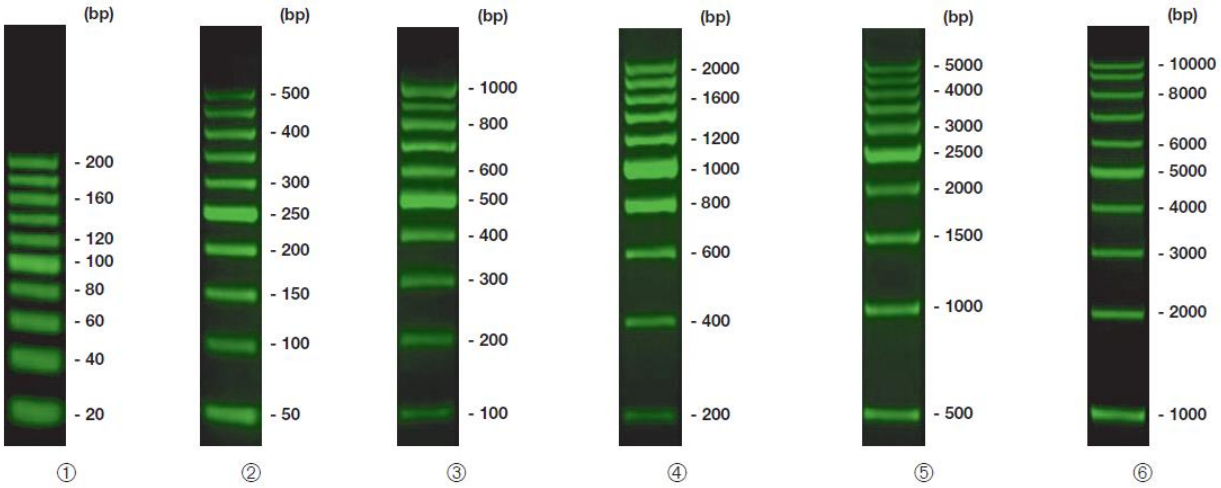
Unevenly Distributed DNA Size Markers

Marker	pUC19-Marker ready-to-use	pUC19/MspI	pBR328-Marker ready-to-use	pBR328 Mix I	Lambda Hind III / phiX Hae III Marker	Lambda Hind III Marker
Art. No.	X901 (fig. 1)	T149 (fig. 2)	X902 (fig. 3)	T146 (fig. 4)	CP49 (fig. 5)	X910 (fig. 6)
Size range (bp)	26 - 501	26 - 501	154 - 2176	154 - 2176	72 - 23130	125 - 23130
Number of fragments	10	10	11	11	17	8
Reinforced band at (bp)	-	-	-	-	-	-
Quantitation	recommended	recommended	recommended	recommended	recommended	recommended
Number of lanes in minigel	200 / ml	200 / 100 µg	200 / ml	200 / 100 µg	200 / 100 µg	200 / 100 µg
Delivery	ready-to-use	lyophilised	ready-to-use	lyophilised	lyophilised	lyophilised



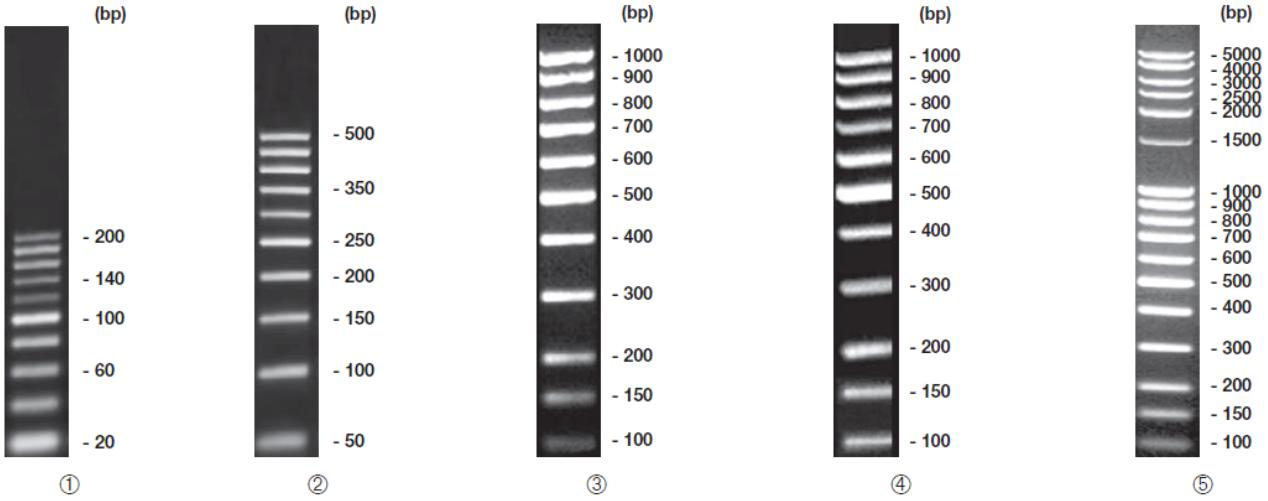
Evenly Distributed DNA Size Markers with Fluorescent Dye

Marker	20 bp-DNA-Fluoro-Ladder	50 bp-DNA-Fluoro-Ladder	100 bp-DNA-Fluoro-Ladder	200 bp-DNA-Fluoro-Ladder	500 bp-DNA-Fluoro-Ladder	1 kbp DNA-Fluoro-Ladder
Art. No.	8262 (fig. 1)	8263 (fig. 2)	8264 (fig. 3)	8265 (fig. 4)	8266 (fig. 5)	8267 (fig. 6)
Size range (bp)	20 - 200	50 - 500	100 - 1000	200 - 2000	500 - 5000	1000 - 10000
Number of fragments	10	10	10	10	10	10
Reinforced band at (bp)	100	250	500	1000	2500	5000
Färbung	fluorescent/green	fluorescent/green	fluorescent/green	fluorescent/green	fluorescent/green	fluorescent/green
Quantitation	possible	possible	possible	possible	possible	possible
Number of lanes in minigel	200 / ml	200 / ml	200 / ml	200 / ml	200 / ml	200 / ml
Delivery	ready-to-use	ready-to-use	ready-to-use	ready-to-use	ready-to-use	ready-to-use

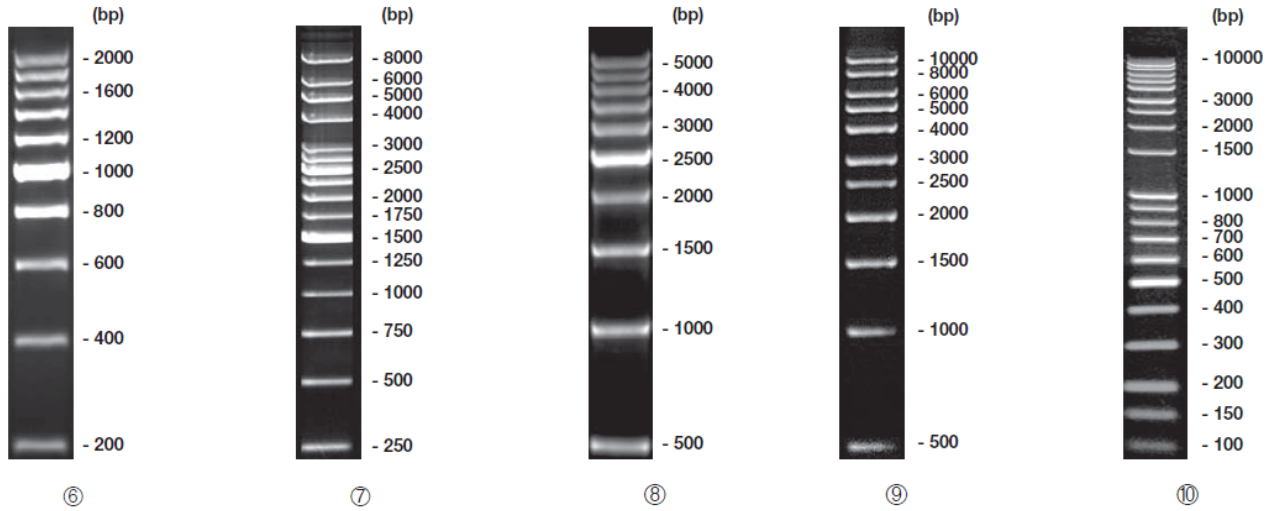


Evenly Distributed DNA Size Markers (Unstained)

Marker	20 bp-DNA Ladder	50 bp-DNA Ladder	100 bp-DNA Ladder equimolar	100 bp-DNA Ladder equalized	100 bp-DNA Ladder extended
Art. No.	2805 (fig. 1)	2810 (fig. 2)	T834 (fig. 3)	T833 (fig. 4)	T835 (fig. 5)
Size range (bp)	20 - 200	50 - 500	100 - 1000	100 - 1000	100 - 5000
Number of fragments	10	10	11	11	17
Reinforced band at (bp)	100	250	500	500	500
Quantitation	possible	possible	recommended	possible	possible
Number of lanes in minigel	200 / ml	200 / ml	200 / 100 µg	500 / 100 µg	160 / 100 µg
Delivery	ready-to-use	ready-to-use	lyophilised	lyophilised	lyophilised

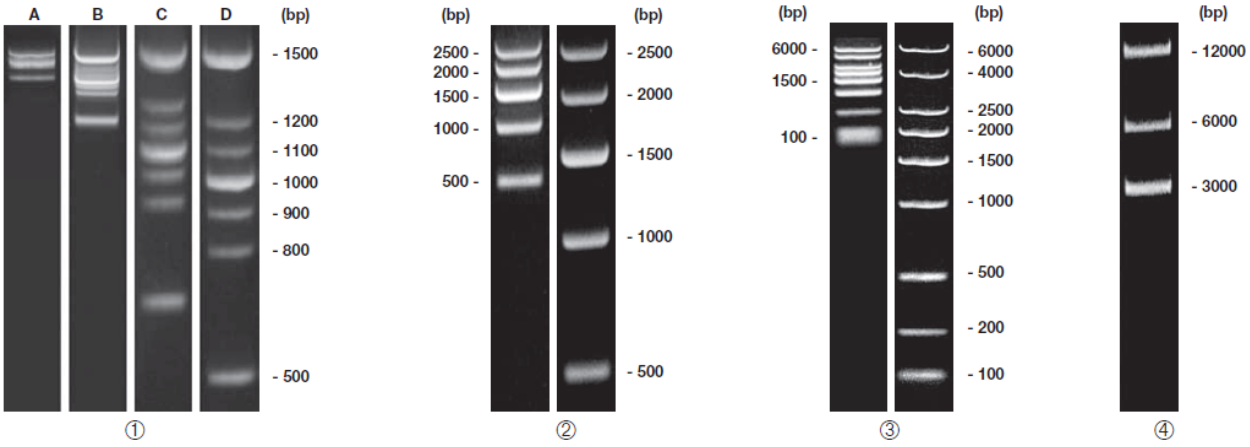


Marker	200 bp-DNA Ladder	250 bp-DNA Ladder	500 bp-DNA Ladder	1 kbp DNA Ladder	DNA Ladder combi
Art. No.	8272 (fig. 6)	T918 (fig. 7)	8273 (fig. 8)	Y014 (fig. 9)	CL22 (fig. 10)
Size range (bp)	200 - 2000	250 - 8000	500 - 5000	500 - 10000	100 - 10000
Number of fragments	10	16	10	11	20
Reinforced band at (bp)	1000	1500, 2500	2500	-	500
Quantitation	possible	-	possible	-	-
Number of lanes in minigel	200 / ml	200 / 100 µg	200 / ml	200 / 100 µg	200 / 140 µg
Delivery	ready-to-use	lyophilised	ready-to-use	lyophilised	lyophilised



DNA Size Markers for Short Runs

Marker	PCR-Marker DNAscore	DNA-Marker short run 1	DNA-Marker short run extended	DNA-Marker ccc-Plasmid
Art. No.	T917 (fig. 1)	0146 (fig. 2)	CL05 (fig. 3)	0149 (fig. 4)
Size range (bp)	500 - 1500	500 - 2500	100 - 6000	3000 - 12000
Number of fragments	7	5	9	3
Reinforced bands at (bp)	500, 1000, 1500	1500	1500	-
Quantitation	possible	possible	possible	-
Number of lanes in minigel	200 / 100 µg	400 / 100 µg	300 / 100 µg	1000 / 100 µg
Delivery	lyophilised	lyophilised	lyophilised	lyophilised



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