



## Contents

Reagents provided are sufficient for 100 applications.

Component	Cat. No. 10488058
TrackIt™ 100 bp DNA Ladder (0.1 µg/µL) <sup>[1]</sup>	500 µL (50 µg)
6X TrackIt™ Cyan/Orange Loading Buffer <sup>[2]</sup>	2 × 500 µL

[1] DNA ladder is provided in buffer consisting of 10 mM Tris-HCl (pH 7.6), 10 mM EDTA, 0.005% xylene cyanol FF, 0.025% Orange G, 10% glycerol.

[2] Loading buffer consists of 10 mM Tris-HCl (pH 7.6), 60 mM EDTA, 0.03% xylene cyanol FF, 0.15% Orange G, 60% glycerol.



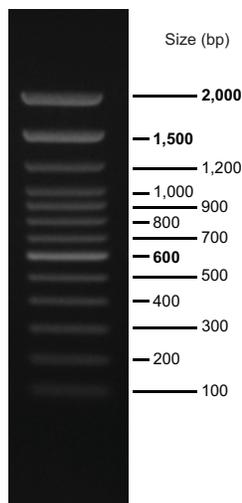
## Storage

- Product is shipped at [ambient temperature](#).
- Store at room temperature or at 4°C for up to 24 months.



## Product description

- The Invitrogen™ TrackIt™ 100 bp DNA Ladder is designed for sizing and quantification of double stranded DNA on 1% to 2% agarose gels.
- The 100 bp DNA Ladder consists of 13 individual chromatography-purified DNA fragments ranging in size from 100 bp to 2,000 bp.
- Three reference bands at 2,000 bp, 1,500 bp, and 600 bp are included for easy orientation.
- The ladder is supplied with 6X TrackIt™ Cyan/Orange Loading Buffer for sample DNA.



## Online resources

- Visit our [product pages](#) for additional information and protocols.
- Go online to view related [DNA ladders and markers](#).
- For support, visit [thermofisher.com/support](http://thermofisher.com/support).



## Required materials

- UltraPure™ Agarose (Cat. No. 16500500)
- Ultrapure™ DNase/RNase-Free Distilled Water (Cat. No. 10977023)
- UltraPure™ TAE Buffer, 10X (Cat. No. 15558042)
- UltraPure™ TBE buffer, 10X (Cat. No. 15581044)
- UltraPure™ Ethidium Bromide, 10 mg/mL (Cat. No. 15585011)
- SYBR™ Safe DNA Gel Stain (Cat. No. S33102)
- SYBR™ Gold Nucleic Acid Gel Stain (10,000X in DMSO) (Cat. No. S11494)



## Important guidelines

- Do not heat the TrackIt™ 100 bp DNA Ladder before loading.
- Load the same volume of DNA sample and DNA ladder.
- For quantification, adjust the concentration of the sample to equalize it approximately with the amount of DNA in the nearest band of the ladder.
- For DNA bands visualization with GelRed™ use gel staining after electrophoresis to avoid aberrant DNA migration.



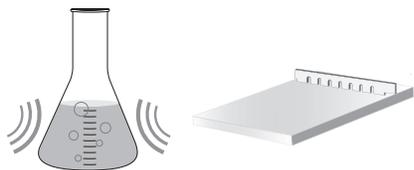
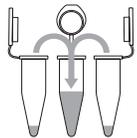
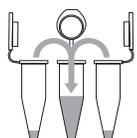
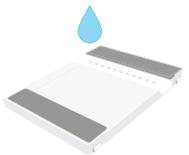
## Guidelines for agarose gel preparation

- Determine the required agarose concentration for your gel based on the size of DNA fragments to be separated.

Fragment size	Recommended agarose gel %	
	1X TAE	1X TBE
800–10,000	0.8	0.7
400–8,000	1.0	0.85
300–7,000	1.2	1.0

- Prepare agarose in a flask with 2-4 times the volume of the agarose solution.
- Exercise caution when handling microwaved agarose. The solution may become superheated and foam over when agitated.
- Refer to the product insert for [UltraPure™ Agarose](#) for detailed instructions on agarose preparation.

## Prepare DNA ladders and samples for electrophoresis

Step		Action												
1		<p><b>Cast agarose gel</b></p> <ol style="list-style-type: none"> <li>Prepare agarose solution (w/v) for the gel percentage appropriate for separating your DNA fragments.</li> <li>Microwave agarose solution.</li> <li>Cast agarose gel.</li> </ol>												
2		<p><b>Prepare DNA ladder</b></p> <ol style="list-style-type: none"> <li>Thaw, mix and briefly centrifuge DNA ladder before use.</li> <li>Mix gently.</li> <li>Load the gel with 1 µL of DNA ladder per 1 mm of well width.</li> </ol>												
3		<p><b>Prepare samples</b></p> <ol style="list-style-type: none"> <li>Dilute your sample with 6X TrackIt™ Cyan/Orange Loading Buffer (Cat. no. 10482028): mix 1 volume of loading dye with 5 volumes of the DNA sample.</li> <li>Mix gently.</li> <li>Load DNA ladder on gel.</li> </ol>												
4		<p><b>Perform electrophoresis</b></p> <ol style="list-style-type: none"> <li>Add appropriate amount of UltraPure TAE or UltraPure TBE buffer to chamber.</li> <li>Set appropriate voltage and perform electrophoresis of samples.</li> </ol> <table border="1" data-bbox="966 852 1963 1023"> <thead> <tr> <th>DNA size</th> <th>Voltage</th> <th>Buffer</th> </tr> </thead> <tbody> <tr> <td>&lt;1 kb</td> <td>5–10 V/cm</td> <td>TBE</td> </tr> <tr> <td>1–5 kb</td> <td>4–10 V/cm</td> <td>TAE or TBE</td> </tr> <tr> <td>&gt;5 kb</td> <td>1–3 V/cm</td> <td>TAE</td> </tr> </tbody> </table>	DNA size	Voltage	Buffer	<1 kb	5–10 V/cm	TBE	1–5 kb	4–10 V/cm	TAE or TBE	>5 kb	1–3 V/cm	TAE
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5		<p><b>Stain agarose gel</b></p> <ol style="list-style-type: none"> <li>Incubate gel in staining buffer for 30 minutes.</li> <li>Visualize DNA ladder and samples. <ul style="list-style-type: none"> <li>Use UV transilluminator to detect DNA bands stained with ethidium bromide.</li> <li>Use blue light transilluminator to detect DNA bands stained with SYBR™ stains.</li> </ul> </li> </ol>												

## Troubleshooting

Observation	Possible cause	Recommended action
Low intensity of all or some DNA bands	Insufficient amount of DNA loaded	Load ~0.1 to 0.2 µg of DNA ladder/marker per 1 mm lane width.
	Insufficient or uneven staining	If DNA will not be used in cloning, add ethidium bromide to both the gel and the electrophoresis loading buffer.
Curved DNA bands	Gel incompletely immersed in electrophoresis buffer	Electrophoresis buffer should completely cover the entire gel during sample loading and run.
	Incorrect electrophoresis conditions	Do not use excessively high voltage electrophoresis.
Atypical banding pattern	Denatured DNA	Excessively high voltage may result gel heating and DNA denaturation.
Smear DNA bands	DNA degradation by nucleases	Use fresh electrophoresis buffers, freshly poured gels, nuclease-free vials and tips.
	Incorrect electrophoresis conditions	Always use the same electrophoresis buffer for both gel and running buffer.
	Poorly formed (slanted) gel wells	When inserting the comb into the gel, make sure that is vertical to the gel surface and stable during gel casting and solidification.
Incorrect quantification data	Incorrect ladder band chosen for sample quantification	Always compare the sample band with a ladder band of similar size.
	Improper quantification method used	If possible use densitometry rather than visual comparison of the bands.
	DNA masking by electrophoresis tracking dye	Do not exceed the recommended amount of electrophoresis tracking dyes used for ladder/sample preparation.

## Guidelines for staining gels

### For staining with ethidium bromide (Cat. No. 15585011)

- Sensitivity of 1 ng/band with UV, and 20 ng/band with blue light transillumination
- Use 0.5 µg/mL of staining dye
- Staining of DNA bands can be performed in gel or after electrophoresis
- For best results of in gel staining use ethidium bromide for gel preparation as well as in electrophoresis running buffer

### For staining with SYBR™ Safe (Cat. No. S33102)

- Sensitivity of 5 ng/band with UV, and 3 ng/band with blue light transillumination
- Staining of DNA bands can be performed in gel or after electrophoresis
- **For staining after electrophoresis:**
- Dilute concentrated SYBR™ Safe staining in 1X TAE or 1X TBE
- Incubate for 30 minutes in dark place under continuous shaking

### For staining with SYBR™ Gold (Cat. No. S11494)

- Sensitivity of 0.1 ng/band with UV, and 0.025 ng/band with blue light transillumination
- Staining of DNA bands can be performed after electrophoresis
- Dilute concentrated SYBR™ Safe staining in 1X TAE or 1X TBE
- Incubate for 10-40 minutes in dark place under continuous shaking

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