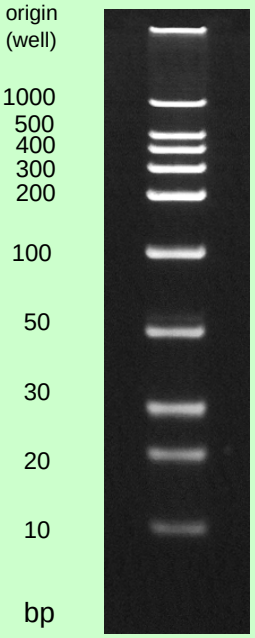
PRODUCT INFORMATION

Product Name : DynaMarker dsRNA
Code No. : DM180
Range : 10 - 1,000 bp of dsRNA
Size : 25 µg (100 µl), 0.25 mg/ml

This product is research use only

Description :

The DynaMarker dsRNA consists of ten double-stranded RNAs, 10, 20, 30, 50, 100, 200, 300, 400, 500 and 1,000 base pairs. The DynaMarker dsRNA is an ideal size marker for determinating sizes of double-stranded RNAs. A twenty bp of RNA band including in the DynaMarker dsRNA is adjusted to approximately 25 ng/µl in concentration. The DynaMarker dsRNA is manufactured for non-denaturing polyacrylamide gel electrophoresis. The DynaMarker dsRNA can be visualized by UV light after ethidium bromide staining.



Electrophoresis profile of DynaMarker dsRNA (0.5 µg), on 7.5 % of polyacrylamide, 1 × TBE buffer as running buffer

Storage buffer :
10 mM Tris-HCl (pH 7.5), 50 mM NaCl , 1 mM EDTA sodium salt

Storage condition :
This product is shipped on dry ice. Upon receipt, store it at - 80°C. Repeated freeze/thaw cycles should be avoided.

Quality Control : After 18 hr incubation of the DynaMarker dsRNA at 37 °C, no visible degradation of the marker is observed in 7.5 % polyacrylamide gel electrophoresis

Note :
Even dsRNA is more resistant to RNase than ssRNA, dsRNA is sensitive to degradation by RNase. To avoid damaging the DynaMarker dsRNA, use care during manipulations to prevent nuclease contamination. Wear gloves and use clean apparatus. Glassware should be pretreated with diethyl pyrocarbonate (DEPC). Nuclease-free disposable plasticware should be used. Solutions and reagents to mix the marker should be high grade and nuclease-free. To use, thaw the DynaMarker dsRNA on ice and keep it on ice while using.

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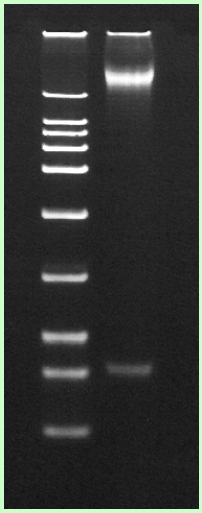
Recommended usage :

The DynaMarker dsRNA is suitable for RNA size determining in non-denaturing polyacrylamide gel electrophoresis. For one of example, DynaMarker dsRNA can be run on 7.5 % polyacrylamide gel as below.

1. Preparation of 7.5 % polyacrylamide gel (20 ml gel)

40 % acrylamide : bis solution (29:1)	3.75 ml
10 × TBE	2.0 ml
H_2O	to 20 ml

2. After mixing reagents described above, add 20 µl of TEMED and 160 µl of 10 % ammonium persulfate. Mix quickly and then pour the gel into the mold of a vertical gel apparatus (20 ml is enough gel solution for two 7 cm × 8 cm, thickness 0.1 cm gels). The gel apparatus should be assembled according to the manufacture's protocol and ready to run with 1 × TBE buffer.



Electrophoresis of DynaMarker dsRNA at the left lane and a long dsRNA digested by dicer at the right lane, on 7.5 % of polyacrylamide, 1 × TBE buffer

3. Loading and electrophoresis

Prepare dsRNA sample for electrophoresis as below.

1) Size Marker:

DynaMarker dsRNA	2 µl (0.5 µg)
nuclease-free water	3 µl
6 × gel loading solution	1 µl

2) Sample to examine:

dsRNA sample	5 µl (50-500 ng)
6 × gel loading solution	1 µl

Mix dsRNA solution with gel loading solution in a tube as above. Load the mixture onto a well of 7.5 % polyacrylamide gel and start electrophoresis. After the tracking dyes have migrated an appropriate distance through gel, stop the electrophoresis. To stain with ethidium bromide, disassemble the apparatus and transfer the polyacrylamide gel to a gel tray filled with 1 × TBE buffer containing 10 µg/ml ethidium bromide. Stained RNA can be visualized using UV transilluminator.

4. Gel loading solution

Gel loading solution is used for non-denaturing polyacrylamide electrophoresis as below.

6 × gel loading solution , example 1
- 0.25 % (w/v) bromophenol blue
- 0.25 % (w/v) xylene cyanol FF
- 30 % (v/v) glycerol in H_2O

6 × gel loading solution , example 2
- 0.25 % (w/v) bromophenol blue
- 40 % (w/v) sucrose in H_2O

Reference:

Sambrook, J. and Russell, D.W. (2001) Molecular Cloning: A Laboratory Manual, 3rd ed., Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.

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