PRODUCT INFORMATION

Product Name: Gel Indicator™ RNA Staining Solution ×100

Code No. : DM595
Size: 10 ml *This product is research use only*

Description: Gel Indicator™ RNA Staining Solution ×100 is 100-fold concentrated solution of Gel Indicator™ RNA Staining Solution (DM590). The ×100 solution is RNase-free. To use the solution, dilute it 100 times with pure water. Gel Indicator™ RNA Staining Solution is developed to stain RNA in polyacrylamide gel and to excise RNA from the gel. The detection limit of RNA is as low as 50 ng. The sensitivity is approximately five times higher than that of UV shadowing. Stained RNA band excised from polyacrylamide gel can be extracted by a crush and soak method followed by ethanol precipitation. Obtained RNA can be used for RT-PCR, enzyme reaction and labeling reaction. Gel Indicator™ RNA Staining Solution does not require a UV transilluminator to detect RNA on polyacrylamide gel because bands are visible under daylight.

Storage condition: Preferable to store at 4°C. Stable at 20° to 25°C for 6 months.
Store prepared 1 × solution at room temperature. This solution should be used within 6 months after dilution.

Quality Control: After 18 hrs incubation of mixture of Gel Indicator™ RNA Staining Solution ×100 and RNA at room temperature (20°-25°C), no visible degradation of RNA is observed in polyacrylamide-urea gel electrophoresis

Note: RNA is very sensitive to degradation by nucleases, although nucleases do not so easily reach to RNA in polyacrylamide gel. To avoid contamination of nuclease, wear gloves and use clean apparatus. Glassware should be pretreated with diethyl pyrocarbonate (DEPC). Nuclease-free disposable plasticware should be used.

Protocol:

1. Dilute Gel Indicator™ RNA Staining Solution ×100 by 100 times with distilled and autoclaved water or nuclease-free ultrapure water to prepare ×1 Solution.
   (For example, add 1 ml of the ×100 solution to 99 ml of ultrapure water)
2. After polyacrylamide gel electrophoresis, transfer acrylamide gel to a tray containing enough volume of distilled autoclaved water (or nuclease-free ultrapure water) to cover the acrylamide gel and rinse the gel for a few minutes.
3. Discard water in the tray and add enough volume of the ×1 Gel Indicator™ RNA Staining to cover the acrylamide gel. Stain the gel for 20-30 min with gentle agitation. RNA will be visible as a blue band. The band can be excised with a razor blade or a gel-excision device.

Caution!
Wear gloves and protective clothing while handling this solution. The stain of Gel Indicator™ RNA Staining Solution is strong.

Ver. 1.0

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Sensitivity of RNA detection:

Gel Indicator™ RNA Staining Solution can detect as low as 50 ng of RNA, even RNA is small.

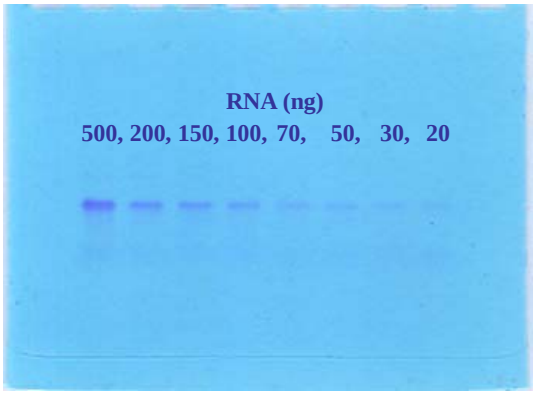

Fig. Staining with Gel Indicator™ RNA Staining Solution

Serially diluted small RNAs (21 base) were subjected to denaturing polyacrylamide gel electrophoresis (12.5 % of polyacrylamide gel containing 7.5 M of Urea, 1× TBE as running buffer) After Electrophoresis, the gel was stained with Gel Indicator™ RNA Staining Solution. It detected 50 ng of 21-base RNA.

Additional Information (Recommended RNA extraction procedure)**:**

RNA integrity after extracted from polyacrylamide gel by the method as below is verified at our lab.

1. Cut excised gel into small pieces with a razor blade and transfer a nuclease-free 1.5-ml tube.
   Excised gel as small as possible, less than 100 µg of gel is preferred to follow this method.
2. Add 3 to 4 volumes of nuclease-free extraction buffer*.
   extraction buffer***:** 0.5 M NH_4OAc, 1 mM EDTA(pH8), 0.1 % SDS
3. Elute the RNA from the gel by agitating vigorously at room temperature for at least 3 hours* (or overnight). *Less than 500-base RNA will be eluted in 3 hours, large-size RNA requires longer incubation time.
4. Add an equal volume of TE-saturated phenol to the mixture of gel and extraction buffer. Vortex it for 15-30 sec.
5. Centrifuge the tube at top speed (15,000 rpm) for 10 min using a microcentrifuge at room temperature. Transfer the supernatant to another nuclease-free 1.5-ml tube.
6. Add equal volume of chloroform to the supernatant. Vortex it briefly. Centrifuge the tube at top speed (15,000 rpm) for 2 min at room temperature. Transfer the supernatant to another nuclease-free 1.5-ml tube. Perform chloroform extraction as above again.
7. Add three volume of 100 % ethanol to obtained supernatant.
8. Put the tube at -80°C over 20 min, then centrifuge the tube at top speed for 10 min. Remove supernatant and add 70 % ethanol. Centrifuge the tube again at top speed (15,000 rpm) for 2 min. Remove the supernatant and dry down the precipitate. Obtained RNA should be dissolved in RNase-free water or buffer depended on your experiment.

Related Products:

DM590	Gel Indicator™ RNA Staining Solution (× 1)		
DM150	DynaMarker RNA Low	DM155	DynaMarker RNA Low Easy Load
DM190	DynaMarker Small RNA	DM195	DynaMarker Small RNA Easy Load

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