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Protocol status: Working
We use this protocol and it's working

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Visualization of RNA using gel electrophoresis method.

khawla alshuraiqi[1]

[1]SQU

khawla alshuraiqi

ABSTRACT

For the analysis of RNA samples, polyacrylamide gel electrophoresis (PAGE) is a highly effective technique. Information about the sample composition and the structural integrity of each unique RNA species is provided by denaturing PAGE. Moreover, Various techniques can be used to see RNA in gels, depending on the type of detection reagent used. Numerous dyes, such as ethidium bromide, SYBR green, and toluidine blue, can be used to stain RNA molecules.

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1 For sample preparation, all samples should be at the same initial concentration by diluting samples with nuclease-free water with a total volume of 9ul and 1 ul for loading dye.

2 Agarose gel preparation

A. Into a flask add 1 ml TAE 50X buffer
B. 49 ml deionized water.
C. 0.75 g agarose powder to make 1.5 % agarose.
D. Heat in the microwave for 1 min, and shake heat again for 1 min.
E. Add the fluorescence dye (Midori Green)
F. Put cast in the sealer.
G. Insert combs according to the number of samples.
H. Let it dry (usually takes 10 min)

3 Running buffer preparation (we can reuse it)

A. From the stock of 50 X prepare 1X by adding 7 ml in 343 ml deionized water.

4 Running the gel

1. Take the gel with its cast and put it in the middle of the tray.
2. Pour the running puffer until it is higher than the wells by 2 mm.
3. Load DNA ladder
4. Mix 9 ul of RNA sample with 1 ul of loading dye using parafilm.
5. Load samples carefully.
6. Cover the gel tray, make sure black to black and red-to-red connections.
7. Connect to the power supply, and run it for 40 min, 80 V.
8. Keep your eyes on the gel to ensure perfect separation.
9. Take the gel to the documentation system.
10. Chose Nucleic Acid gel settings, and capture.
11. There should be 2 clear bands one at 28 (thick band) Twice intense as the other band, and the other at 18 (thinner band).