## **INSTRUCTIONS**



## GeneGelRed™

### **Nucleic Acid Gel Stain**

#### Package Contents: 1 vial

GeneGelRed <sup>™</sup> Nucleic Acid Gel Stain					
Catalog No.	Package	Price	Quantity/Unit	Form	Sipping and Storage Guidelines
R-GR01	0.5 ml	90	- 1 vial.	Liquid. Deep red.	Stable for room temperature. Recommended storage at 4°C or - 20°C,effective for 1year.
R-GR02	1 ml	144			
R-GR03	5 ml	576			
R-GR04	10 ml	1050			

#### Storage

Stable at room temperature. Recommended storage at 4°C or -20°C.

#### DESCRIPTION

GeneGelRed is the best nucleic acid gel stain for its sensitivity, safety and stability. It represents a new generation of fluorescent nucleic acid dye designed to replace the highly toxic ethidium bromide (EB) for staining dsDNA, ssDNA or RNA in agarose gels or polyacrylamide gels. It is far more sensitive than EB requiring no destaining step. Considering GeneGelRed and EB have the same spectra virtually, researchers can directly replace EB with GeneGelRed without changing the instrument setting used for EtBr (Figure 1).

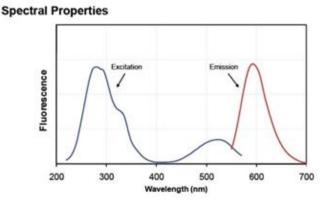


Figure 1. Excitation and emission spectra of GeneGelRed.



GeneGelRed can be used to stain dsDNA, ssDNA or RNA and is highly sensitive either as precast gel stains or post gel stains. To stain dsDNA, ssDNA or RNA in polyacrylamide gels, post gel staining is required. GeneGelRed is compatible with downstream DNA manipulations such as restriction digest, sequencing, and cloning.

Nucleic acid dyes are inherently hazardous due to their potential to cause DNA mutation, while GeneGelRed is unable to cross cell membranes, thus denying the opportunity for them to interact with intracellular DNA. In addition, extensive tests, including those by third party labs, have confirmed that GeneGelRed is noncytotoxic, nonmutagenic and nonhazardous for disposal.

#### **ADVANTAGES**

1. Safer than EB: Non-mutagenic and noncytotoxic proved by Ames and other tests.

2. Easy disposal: GeneGelRed has passed environmental safety tests for direct disposal down the drain or in regular trash.

3. Ultra-sensitive: Much more sensitive than EtBr and other EtBr alternatives.

4. Extremely stable: Available in water, stable at room temperature for long-term storage and microwavable.

5. Simple to use: Simple procedures for precast or post-electrophoresis gel staining.

6. Perfect compatibility with a standard UV transilluminator ore a Gel Reader, which makes GeneGelRed replace EtBr with no optical setting change.

7. Flexible for different downstream applications: Gel purification, restriction digest, sequencing and cloning.

#### PROCEDURE

#### **Post-Staining Protocol**

- 1. Run gels according to your standard protocol.
- 2. Dilute the 10,000× stock and make a 3× staining solution in H<sub>2</sub>O. 50 mL staining solution is adequate for one minigel.

#### Note:

- 0.1 M NaCl in the staining solution enhances sensitivity, but may promote dye precipitation if the gel stain is reused.
- 3. Place the gel in a suitable container and submerge the gel with the  $3 \times$  staining solution.
- 4. Agitate the gel gently at room temperature for 30 min.



#### Note:

Optimal staining time may vary, somewhat depending on the thickness of the gel and the percentage of agarose. For polyacrylamide gels containing 3.5-10% acrylamide, typical staining time is 30 min to 1 h. For gels of higher acrylamide content stain longer.

- 5. Although destaining is not required, the gel can be washed in water to reduce background.
- 6. View the stained gel with a standard transilluminator (302 or 312 nm) and image the gel using an ethidium bromide filter. SYBR or GelStar filters also may be used for gel imaging with equally good results.

#### Note:

Staining solution can be reused at least 2-3 times. Store staining solution at room temperature protected from light.

#### **Precast Protocol for Agarose Gels**

1. Prepare molten agarose gel solution using your standard protocol.

#### Note:

The precast protocol is not recommended for polyacrylamide gels. They can be stained according to the post-stain protocol.

2. Add 10,000× stock reagent into the molten agarose gel solution at 1:10,000 dilution and mix thoroughly.

#### Note:

GeneGelRed can be added while the gel solution is still hot.

- 3. Cast the gel and allow it to solidify.
- 4. Load samples onto the gel and run the electrophoresis using your standard protocol.
- 5. View the stained gel and image the gel.

*Note:* Unused agarose containing GeneGelRed can be remelted to cast more gels, but it may be necessary to add more dye for optimal signal. Do not store agarose containing GeneGelRed in molten form (i.e., at 50 °C) for more than a few days.

Precast gels containing GeneGelRed can be stored for up to a week. Store gels at room temperature in the dark. Storage of GeneGelRed precast gels at 4 C can cause dye precipitation and poor performance.

#### CAUTIONS

1. Post-staining of gels is highly recommended (high affinity of nucleic acid binding dyes can affect DNA migration during electrophoresis).

2. The precast protocol is not recommended for polyacrylamide gels.

3. GeneGelRed can be used to stain dsDNA, ssDNA or RNA. GeneGelRed is twice more sensitive for dsDNA than



4. Gel staining with GeneGelRed is compatible with downstream applications such as sequencing and cloning.

5. GeneGelRed can be removed from DNA by phenol/chloroform extraction and ethanol precipitation.

#### TROUBLESHOOTING

#### 1. Why are the DNA band(s) smeared or smiling or discrepant DNA migration?

Many customers use GeneGelRed precast gels for convenience. However, because GeneGelRed is a high affinity dye designed to improve their safety, it can affect the migration of DNA in precast gels. Some samples, such as restriction digested DNA may migrate abnormally in GeneGelRed precast gels.

Tip 1: Load less DNA: Smearing and smiling in GeneGelRed precast gels are most caused by overloading of DNA. If you see band migration shifts or smearing and smiling, try to reduce the amount of DNA loaded. The recommended loading amount for ladders and samples of known concentration is 50-200 ng/lane. For samples of unknown concentration, try loading one half or one third of the usual amount of DNA.

Tip 2: Try the post-staining protocol: To avoid any interference the dye may have on DNA migration, we recommend using the post-staining protocol. If your application requires loading more than the recommended amount of DNA, use the post-staining protocol. Though we recommend post-staining gels for 30 minutes, you may be able to see bands in as little as five minutes, depending on how much DNA is present. Post-staining solutions can be reused.

Other tips to improve agarose gel resolution:

If DNA migration issues or smearing after post-staining with GeneGelRed appear, and the problem of the nucleic acid dye is excluded, try to avoid overfilling gel wells.

Pour a lower percentage agarose gel, for higher molecular weight DNA separates better with a lower percentage gel. Change the running buffer, for TBE buffer has a higher buffering capacity than TAE buffer.

# 2. Why do I see weak fluorescence, decreased dye performance over time, or a film of dye remaining on the gel after post-staining?

There are a few possibilities:

A. The dye may have precipitated out of solution. Heat the GeneGelRed solution to 45-50°C for two minutes and vortex to dissolve. Store dye at room temperature to avoid precipitation.

B. If you are seeing high background staining of the gel, the agarose that you are using may be of low quality. Contaminants in the agarose may bind to the dye, resulting in increased background.



### **Contact Information**

Genemedi Biotech. Inc.

For more information about reagents, please visit: <u>https://www.genemedi.net/i/reagent</u> For more information about Genemedi products and to download manuals in PDF format, please visit our web site: <u>www.genemedi.net</u> For additional information or technical assistance, please call or email us

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