

November 12, 2018

Product Information

GelRed® Prestain Plus 6X DNA Loading Dye

Catalog Number: 41011

Unit Size: 1 mL

Storage and Handling

Store at 4°C, protected from light. Product is stable for at least 6 months from date of receipt when stored as recommended. While GelRed® has undergone extensive safety testing, Biotium recommends following universal safety precautions when working in the laboratory.

Product Description

GelRed® is a sensitive, stable and environmentally safe fluorescent nucleic acid dyes designed to replace the highly toxic ethidium bromide (EtBr). GelRed® Prestain Plus 6X DNA Loading Dye contains density agents, tracking dyes, and GelRed® dye. The 6X prestain loading dye is added to samples in place of gel loading buffer, and eliminates the need to add fluorescent DNA dye to the agarose gel during casting or after electrophoresis. The loading dye contains two blue electrophoresis tracking dyes that run at approximately 1.5 kb and 200 bp in a 1% agarose gel.

GelRed® Prestain Plus 6X DNA Loading Dye is an improved version of our original 6X GelRed® Prestain Loading Buffer (catalog number 41009) with brighter signal and more consistent DNA migration. When DNA is bound to GelRed® before electrophoresis, the ratio of dye to DNA can cause variable shifts in DNA migration, making it difficult to compare DNA fragment sizes between samples. GelRed® Prestain Plus 6X DNA Loading Dye is formulated to minimize this DNA migration shift, for greater consistency. GelRed® precast gels. See frequently asked questions, next page, for a comparison of GelRed® staining protocols.

GelRed® and EtBr have virtually the same spectra (Figure 1), so you can directly replace EtBr with GelRed® without changing your existing imaging system. In addition, GelRed® is far more sensitive than EtBr, which cannot be used in DNA loading buffer to prestain DNA. GelRed® is compatible with downstream applications such as sequencing and cloning. It is efficiently removed from DNA by gel extraction kits or by phenol/chloroform extraction and ethanol precipitation.

GelRed® Safety

GelRed® was subjected to a series of tests at Biotium and by three independent testing services to assess the dye's safety for routine handling and disposal. Test results confirm that the dye is impenetrable to both latex gloves and cell membranes. The dye is noncytotoxic and nonmutagenic at concentrations well above the working concentrations used in gel staining. GelRed® successfully passed environmental safety tests in compliance with CCR Title 22 Hazardous Waste Characterization, under which the dye is not classified as hazardous waste. A complete safety report is available at www.biotium.com.



Figure 1. Excitation and emission spectra of GelRed® dye in the presence of dsDNA.

Product Protocol

- Prepare agarose gel according to your standard protocol. Do not add ethidium bromide, GelRed®, or any other fluorescent DNA dye to the agarose or buffer.
- Briefly vortex GelRed® Prestain Plus 6X DNA Loading Dye. Add the dye to your DNA samples at a volume ratio of 1:5 (for example, mix 10 uL sample + 2 uL dye).
- 3. Load samples and run gels according to your standard protocol.

CalRad® Prostain Plus

 Visualize bands using a UV transilluminator or other gel documentation system. Gels can be imaged using an ethidium bromide emission filter. SYBR® Green or GelStar™ filters also can be used for gel imaging with equally good results.

Original ColRod® 4V Prostain

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1 kb DNA Ladder (* 1000 bp band, 60 ng)



Figure 2. Comparison of GelRed® Prestain Plus 6X DNA Loading Dye and original 6X GelRed® Prestain Loading Buffer. Two-fold dilutions of 1 kb or 100 bp DNA ladder were mixed with the indicated loading buffer and run on 1% agarose/TBE gels. Total ng of DNA loaded per lane are indicated. GelRed® Prestain Plus shows brighter signal and more consistent band migration compared to original 6X GelRed® Prestain. Table 1. Comparison of GelRed® staining methods

Method / Product	Catalog No.	Procedure	Advantages	Disadvantages	Recommended for
Sample prestaining with GelRed® Prestain Plus	41011	GelRed® loading buffer is added directly to the DNA sample before loading	 Fast & simple: one-step sample loading & DNA staining Less concentrated dye for safer handling Can re-run a gel to use empty lanes 	Not recommended for PAGE, DGGE, EMSA, or PFGE gels	 Routine agarose gels Recommended loading 50-200 ng ladder or 2-5 uL PCR product (~100 ng/ band or less)
Precast gel staining with GelRed® 10,000X in water	41003	GelRed® is mixed with molten agarose before gel casting	Familiar protocol, rapid results	 Dye may cause band migration issues when loading larger amounts of DNA (more than ~100 ng/band), or for some restriction digests 	
Precast gel staining with GelRed® Agarose LE	41029	Agarose is supplied pre- coated with GelRed®, just dissolve, heat, and pour	Safer & more convenient, no need to handle concentrated dye		
Post- electrophoresis gel staining with GelRed® 10,000X in water or GelRed® 3X in water	41003 or 41001	No fluorescent dye is added to the gel, it is stained in 3X GelRed® solution after electrophoresis	 Most accurate sizing/sharpest bands Staining solution can be re-used Enhance sensitivity by adding NaCl 	Extra staining step (up to 30 minutes) after electrophoresis (some customers report good results after only 5 minutes if dye is not reused)	 Highly accurate band sizing If more than ~100 ng DNA per band must be loaded Analyzing restriction digests

Frequently Asked Questions	Answers
Can GelRed® stain RNA or ssDNA?	GelRed® can be used to stain ssDNA and RNA, but it is twice as sensitive for dsDNA than for ssDNA or RNA.
Is GelRed® compatible with cloning, ligation and sequencing?	Yes. Gel extraction or phenol-chloroform extraction can be used to remove dye from DNA. Biotium's gel extraction kit has been validated for removal of GelRed® and GelGreen®, as well as kits from other suppliers.
Can GelRed® be used for formaldehyde, polyacrylamide, DGGE, EMSA or PFGE (pulse-field) gels?	Yes. Customers have reported using GelRed® in glyoxal and formaldehyde agarose gels for precast staining of RNA. Use the post-staining protocol for polyacrylamide, DGGE, EMSA, and PFGE gels.
Is GelRed® compatible with Southern or northern blotting?	GelRed® has been validated for Southern blotting (Plant Cell Report <u>doi:10.1007/s00299-011-1150-7</u>). We recommend using the post-staining protocol for blotting applications.
How should I dispose of GelRed®?	GelRed® has passed the EPA regulated Title 22 test. Some facilities have approved the disposal of GelRed® directly down the drain. However, because regulations vary, please contact your safety office for local disposal guidelines. If required, GelRed® can be adsorbed by activated charcoal bags (catalog no. 22007) for disposal as chemical waste.
What is the lower detection limit of GelRed®?	Some users have reported being able to detect bands containing less than 0.1 ng DNA. However, the limit of detection will depend on instrument capability and exposure settings.
What is the binding mechanism of GelRed®?	GelRed® most likely binds by a combination of intercalation and electrostatic interaction.
What is the chemical structure of GelRed®?	The chemical structure of GelRed® is proprietary.
Does GelRed® migrate during electrophoresis?	GelRed® does not migrate through the gel as easily as EtBr. It is not necessary to add dye to the running buffer, and the gel will be stained more homogeneously with GelRed® than with EtBr.
I accidentally left my GelRed® in the light. Will it still work?	While we recommend that you protect the dye from light during long term storage, we have had a customer report using GelRed® with success after accidentally leaving it in ambient light for one month.

Related Products

Catalog number	Product
41028	Agarose LE, Ultrapure Molecular Biology Grade
41029	GelRed® Agarose LE
41006	TBE Buffer, 5X (4L Cubitainer®)
99962-1	6X DNA Loading Buffer (Blue)
31022	Ready-to-Use 1 kb DNA Ladder
31032	Ready-to-Use 100 bp DNA Ladder
22007	Activated Charcoal Decontamination Bags
31030	DNA Gel Extraction Kit
41001	GelRed® Nucleic Acid Gel Stain, 3X in Water
41003	GelRed® Nucleic Acid Gel Stain, 10,000X in Water
41005	GelGreen® Nucleic Acid Gel Stain, 10,000X in Water
41020	DNAzure® Blue Nucleic Acid Gel Stain
E90003	Gel-Bright™ LED Gel Illuminator

Please visit our website at <u>www.biotium.com</u> for information on our life science research products, including environmentally friendly EvaGreen® qPCR master mixes, fluorescent CF® dye antibody conjugates and reactive dyes, apoptosis reagents, fluorescent probes, and kits for cell biology research.

GelRed is covered by US and international patents. Cubitainer is a registered trademark of Hedwin Corporation GelStar is a trademark of FMC Corporation. SYBR is a registered trademark of Thermo Fisher Scientific.

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