vivantis

DYE PRODUCT

Product Datasheet

ViSafe Green Gel Stain (10000X in water)

Product No : SD0101
Quantity : 500µl/pack

Safe Green

10000X in H_2O

Lot :
Expiry Date :
Concentration : 10000X

Shipped at ambient temperature
Store at 2-8°C or -20°C

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DESCRIPTION

ViSafe Green Gel Stain is a stable, sensitive and environmentally safe fluorescent nucleic acid dye for staining double-stranded DNA (dsDNA), single-stranded DNA (ssDNA) or RNA in agarose gels or polyacrylamide gels.

ViSafe Green Gel Stain has UV absorption between 250nm and 300nm. It is compatible with either 254nm UV transilluminator or a gel reader equipped with visible excitation. Both green gel stain and EtBr have the same spectra, so the green gel stain able to replace Ethidium Bromide (EtBr) without changing existing imaging system.

The green gel stain is designed to replace the highly toxic ethidium bromide (EtBr). The dye is confirmed by Ames test results that it is impenetrable to latex gloves and cell membranes. By using the suggested working concentrations in gel staining, the dye is proven unable to cross cell membranes; and it is noncytotoxic and nonmutagenic at working concentrations.

ViSafe Green Gel Stain, 10000X in H_2O, can be diluted 10000X for precast gel protocol or 3000-3300X for post gel staining. One vial (0.5ml) of 10000X solution can be used for at least 100 minigels either using precast method or post-staining method.

FEATURES

Safer
The stain is noncytotoxic & nonmutagenic shown by Ames tests.

Higher sensitivity
More sensitive compared to EtBr or Viva SybrGreen Nucleic Acid Stain.

Extremely stable
Stable at room temperature for long-term storage. Stable to be microwaved or being heated. The working solution is stable at room temperature when kept in dark.

Wide application
Suitable to stain dsDNA, ssDNA and RNA. Suitable to use in agarose gel or polyacrylamide gel. Compatible with downstream applications, such as gel recovery & cloning.

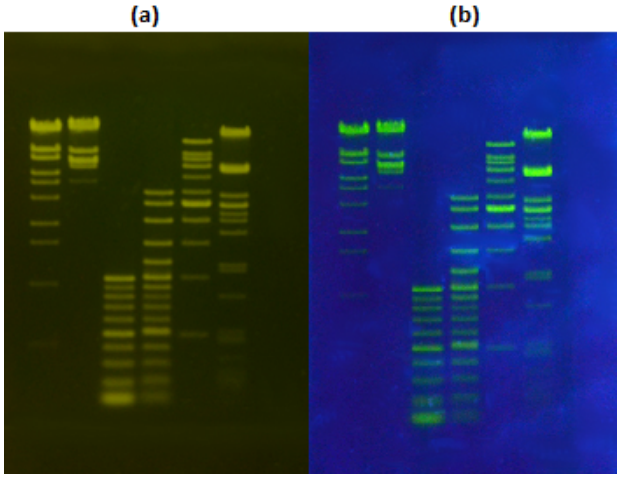
Easy staining protocols
Easy precast gel staining & post-staining procedures.

Compatible with most imaging system
Gel can be viewed with standard UV transilluminator, visible light gel reader or other gel imaging system.

STAINING PROTOCOL

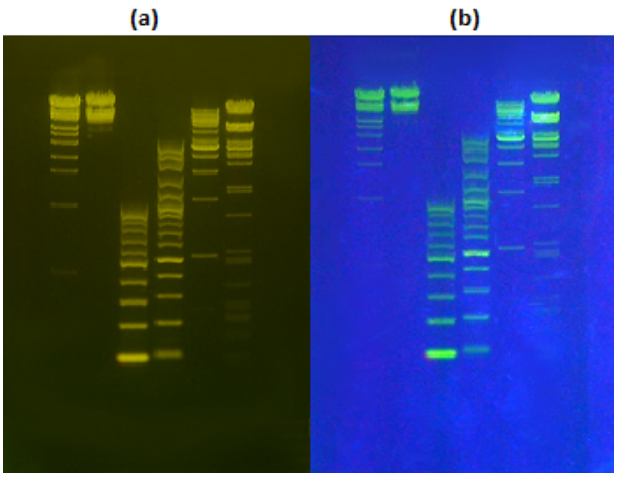
Due to ViSafe Green Gel Stain's slow diffusion rate in the relatively tight polyacrylamide gel matrix, the green gel stain is not suggested for staining DNA or RNA in precast polyacrylamide gels. Post-staining method can be used for polyacrylamide gels.

Using precast gel with green gel stain is more convenient. However, some DNA samples may experience migration retardation or compromised resolution in the presence of green gel stain. The green gel stain may affect DNA migration during electrophoresis. Hence, post-staining with green gel stain is highly recommended. Post-staining method may lead to gel results with higher sensitivity and without dye interference with DNA migration. Besides, post-staining method is a simple protocol with no destaining and no special buffer needed.



Post-staining for Agarose Gel

Figure 1: Various ladders and makers run at 1.5% TBE agarose gel. The agarose gel is post-stained with ViSafe Green Gel Stain. The gel is visualized using transilluminator with (a) blue light; (b) UV light.



Precast for Agarose Gel

Figure 2: The agarose gel is pre-stained with ViSafe Green Gel Stain. Various ladders and makers run at 1.5% TBE pre-stained agarose gel. The gel is visualized using transilluminator with (a) blue light; (b) UV light.

Precast Protocol for 1% Agarose Gel

1. Pour 1g of agarose powder and 100ml 1X TAE or 1X TBE into glass flask.
2. Melt the agarose in microwave for 1-3 mins until the agarose is completely dissolved.
3. Add 5µl green gel stain per 50ml gel and stir the gel solution to mix thoroughly.
   *Dilute the stock solution into agarose gel solution at 1:10000.
   *Since the green gel stain is thermally stable, the stock solution can be added while the gel solution is still hot.
   *The green gel stain can be pre-combined with agarose powder and gel working solution followed by microwaving or other heating procedures. Make sure the green gel stain is swirled and stirred well to mix with gel solution.
4. Pour the agarose solution onto gel plate and insert a comb.
5. Place newly poured gel at 4°C for 10-15 mins or stay at room temperature for 20-30 mins, until it has completely solidified.
   *ViSafe Green Gel Stain precast gels can be prepared in larger quantities and stored for later use since the stain is hydrolytically stable. Any leftover gel solution can be stored and re-heated for additional gel casting. Avoid heating the green gel stain gel solution more than 3 times.
6. After the gel is ready, perform gel electrophoresis.
7. Visualize or image the gel directly under UV light or blue light after gel electrophoresis is done.

Post-staining Protocol for Agarose Gel and Polyacrylamide Gel

1. Run the gel electrophoresis accordingly.
2. Dilute 10000X ViSafe Green Gel Stain stock solutions to 3X staining solutions with 0.1M NaCl water solution. (This solution can be used at least 2-3 times,protected from light. Suggested to use container covered with aluminium foil or use dark colour container.)
   Example: **30µl of green gel stain added into 100ml of 0.1M NaCl solution**
   *NaCl solution in the staining solution is optional. Adding NaCl in the staining solution enhances the staining, but may promote dye precipitation if the staining solution is to be used repeatedly. Any staining solution to be reused is preferably stored at room temperature in a dark place to reduce possible dye precipitation problem.
3. Remove the gel from the gel tank and transfer into staining container.
4. Allow the gel to stain for at least 25-30 mins with gentle shaking.
5. Destaining is not necessary, but rinsing the gel with water can reduce the background.
6. Visualize or image the stained gel under UV light or blue light.

TIPS

1. To increase resolution of the DNA bands:
   a. Running the gel at a lower voltage for longer period of time
   b. Using a wider gel comb
   c. Loading less DNA into well
2. To get better separation of bands, adjust the agarose percentage of the gel if the similarly sized bands that that are running too close together are loaded. A higher percentage agarose gel will help in resolving smaller bands from each other, and a lower percentage gel will help in separating larger bands.
3. ViSafe Green Gel Stain is efficiently removed from DNA by phenol/chloroform extraction and ethanol precipitation.

Smeared bands observed or poor band separation in precast gel

1. Post-staining is suggested to perform.
2. Reduce DNA amount loaded per well.
3. Lower percentage of agarose gel for large fragments for better resolution.
4. Run full length or longer gel.
5. Increase gel solidification duration to have sharp formation.
6. Change suitable running buffers. TBE buffer has higher buffering capacity compared to TAE buffer.
7. Polyacrylamide gel is not recommended for precast or pre-staining method.

Inconsistency of DNA migration in precast gel

1. Reduce DNA amount loaded per well.
2. Reduce dye amount used.
3. Post-staining is suggested because the dye will not be interfered with DNA migration during electrophoresis.

Dye or fluorescence performance decreases over time

1. Precipitation may occur in solution. Heat the safe dye solution to 45-50°C for two minutes and swirl or stir the solution well.
2. Store the safe dye solution at room temperature and keep in dark.

RECOMMENDED EQUIPMENT REQUIRED BUT NOT SUPPLIED

1. ElitePro Single Channel Adjustable Volume Pipette, 1-10uL (Hercuvan Cat No: TT-EP-10U)
2. MiniGEL Electrophoresis System (Hercuvan Cat No: TT-HES-1-110V/TT-HES-1-220V)
3. SafeViewer Blue Light Transilluminator (Hercuvan Cat No: TT-BLT-470)

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