CYANAGEN

Reagents for Genomics and Proteomics

NEW

THE RIGHT LIGHT

GREEN STAIN

non-hazardous fluorescent stain
for nucleic acid detection in gels
(10.000 X in DMSO)

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About us

Cyanagen is a biotech company located in Bologna, dedicated to research, development and production of reagents for molecular diagnostic since 2003 and one of the leading companies in the field of reagents for Western blotting and Elisa.
The main product lines are focused on chemiluminescence and fluorescent dyes for biological analysis, genomics, proteomics and chemical sensors.
They are based on Cyanagen internationally patented technologies and achieve outstanding performance in terms of sensitivity and stability.
The products are extremely versatile and perfectly suited to the latest analytical instrumentation. These products are also available as OEM.

Cyanagen s.r.l. has a certified Quality System

ISO 9001:2015 – ISO 13485:2016

Product manual

Green Stain

Non-hazardous fluorescent stain for nucleic acid detection in gels
(10000X in DMSO)

GREEN STAIN IS INTENDED FOR RESEARCH USE ONLY AND SHALL NOT BE USED IN ANY CLINICAL PROCEDURES OR FOR DIAGNOSTIC PURPOSES.

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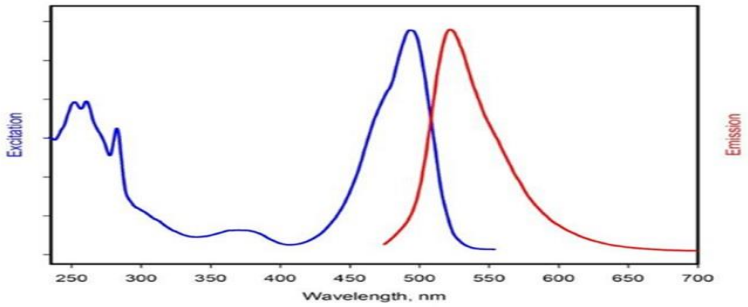
1. Introduction

Nucleic acid gel staining has been performed with ethidium bromide for decades. Even though ethidium bromide is very economic and easy to use, its mutagenicity and hazardous waste disposal fueled the development of DNA gel staining alternatives. Cyanagen has developed Green Stain to solve these issues. Green Stain has a high binding affinity for nucleic acids providing bright green light-emitting DNA-dye complexes. Bright green fluorescent bands and very low background fluorescence are its major features. In addition, Green Stain has a very low toxicity combined with an extraordinary sensitivity. It can be used for any gel staining application requiring the detection of up to 100 pg of genomic dsDNA with sufficient sensitivity. Green Stain preferentially binds double-strand DNA, it also allows also single-strand DNA and RNA detection, although with lower sensitivity. Green Stain is provided as a concentrate solution that can be used with the same protocols applied to ethidium bromide. Green Stain can be used with pre-, post- electrophoresis gel staining and DNA sample pre-staining. The nucleic acid-bound Green Stain is efficiently excited at **~254 nm** and **~488 nm.** Detection can be performed with the same instruments used for ethidium bromide and SYBR® Green gel staining such as standard UV transilluminator (254 nm) as well as with CCD-camera imaging system or laser-based scanner selecting the SYBR® Green filter.



Excitation/emission spectra of Green Stain bound to dsDNA

Features:

- Very low toxicity;
- Ready to use kit;
- High sensitivity: detection up to 100 pg per band;
- Optimal signal to background ratio;
- Simple substitution of your stain reagent with Green Stain.

Storage

- Store at – 20°C;
- Avoid repeated freeze-thaw cycles;

Follow universal safety precautions.

2. Components and other materials required

Kit components

- One package includes 1 x 500µl or 2 x 500µl Green Stain concentrate in DMSO.

Other materials required

- TE (10 mM Tris-HCl, 1 mM EDTA, pH 7.5), TBE (89 mM Tris base, 89 mM boric acid, 1 mM EDTA, pH 8) or TAE (40 mM Trisacetate, 1 mM EDTA, pH 8) buffer
- Agarose

3. Staining protocols

Green Stain can be used in different staining protocols: pre-electrophoresis gel staining, post-electrophoresis gel staining, and sample pre-staining. Since Green Stain has high affinity for nucleic acids, the binding of the dye can affect DNA/RNA migration during electrophoresis. To avoid any interference, we recommend to use the sample pre-staining protocol.

1. **Pre-electrophoresis gel staining:**
   This method is acceptable for agarose gels only, but not for nondenaturing polyacrylamide gel.
   Both TBE and TAE buffers are compatible with this procedure.
   Wear clean powder free gloves during all gel handling.
   - Dissolve the agarose in the buffer using microwave or heating appliance.
   - Thaw Green Stain at room temperature
   - Add Green Stain to the gel solution (1:10000 dilution). Mix thoroughly.
   - Pour the gel and let it cool down.
   - Perform electrophoresis in an agarose gel in TBE or TAE buffers according to standard procedures.
   - Visualize or document the gel
2. **Post-electrophoresis gel staining:**
   Wear clean powder free gloves during all gel handling
   - Perform electrophoresis in TBE or TAE buffers according to standard procedures.
   - Thaw Green Stain at room temperature and dilute 1:2000 in TE, TBE, or TAE buffers.
   - Place the gel in the staining solution and incubate at room temperature for 15–40 minutes with gentle agitation. Use a plastic container and protect the staining solution from light. Always wrap the staining container in aluminum foil to protect from direct light. Staining containers should be meticulously clean.
   - Visualize or document the gel

3. **Sample pre-staining:**
   - Dilute Green Stain 1:100 in TE or TBE buffer.
   - Use the Green Stain working solution as 2X and add it to the DNA sample plus loading dye and incubate at least 5 min.
   - Perform electrophoresis in TBE or TAE buffers according to standard procedures.

- Visualize or document the gel

Gel visualization can be done with a standard transilluminator (254 nm), with a CCD-camera imaging system or with a laser-based scanner selecting the SYBR® Green filter.

NOTE: if you are not satisfied with your results and you have some problems as poor efficiency, distorted migration etc.., please refer to troubleshooting.

4. Troubleshooting

Poor staining efficiency

Possible Cause	Precautions/Remedies
Reduced concentration of the stock solutions	During incomplete thawing, Green Stain is more concentrated in the first thawed volume. Repeated cycles of incomplete thawing will decrease the concentration of the remaining stock solution. Always achieve complete thawing before pipetting Green Stain.
Repeated thaw-freeze cycles	Divide the Green Stain stock solution into aliquots.
SDS contamination	Avoid the presence of SDS in the staining solution, in the loading dye or in the running buffer. Do not wash staining container with detergent solutions.
pH of the buffer solution	pH of the buffer solution must range from 7.5 to 8.3.
Imaging system	Use only transilluminator with 254 nm UV-Lamp or Led/Laser based imaging instruments at 460-488 nm.

Discrepant or distorted DNA migration

Possible Cause	Precautions/Remedies
DNA overloading	Prefer the sample pre-staining protocol. When using pre-electrophoresis gel staining, reduce the amount of DNA loaded (for example, each lane should contain less than 50 ng of DNA ladders)
Staining protocol	If you cannot reduce the amount of loaded DNA, use the sample pre-staining protocol
Density of agarose gel	Reduce the percentage of agarose in the gel
High voltage	Reduce voltage to avoid localized heating of the gel resulting in aberrant migration of bands or even heat-caused damage to the dyes

Exhausted running buffer	Change the running buffer with a freshly prepared buffer. Since TAE has a lower buffer capacity, use TBE buffer

5. Ordering information

Product Description:	Quantity:	Sufficient For:	Order-No:
Green Stain 10000 X in DMSO	0.5 mL	50 minigels stain	NAGS068,0005
	2 x 0.5 mL	100 minigels stain	NAGS068,0010

For further information visit **www.cyanagen.com**

For orders: call **+39 051.534063**
mail to **sales@cyanagen.com**

Warranty Disclaimer at
www.cyanagen.com/warranty-disclaimer/

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GREEN STAIN

Cyanagen srl has a certified Quality System

ISO 9001:2008 QUALITY CERTIFIED

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