SYBR Safe™ DNA Gel Stain

The safer ethidium bromide alternative

Quick Facts

Storage upon receipt:

- Room temperature

Ex/Em: 280, 502/530 nm, bound to DNA

Introduction

SYBR Safe™ DNA gel stain has been specifically developed for reduced mutagenicity, making it safer than ethidium bromide for staining DNA in agarose or acrylamide gels. SYBR Safe stain comes either as a concentrate or as a ready-to-use solution that can be used just like an ethidium bromide solution, and the detection sensitivity with SYBR Safe stain is better than with ethidium bromide. DNA bands stained with SYBR Safe DNA gel stain can be detected using a standard UV transilluminator, a visible-light transilluminator or a laser-based scanner. The stain is also suitable for staining RNA in gels. Bound to nucleic acids, SYBR Safe stain has fluorescence excitation maxima at 280 and 502 nm, and an emission maximum at 530 nm (Figure 1).

Materials

Contents

SYBR Safe DNA gel stain is supplied ready-to-use in 0.5X TBE buffer (45 mM Tris-borate, 1 mM EDTA, pH ~8.3; S33100, S33101) or 1X TAE buffer (40 mM Tris-acetate, 1 mM EDTA, pH ~8.3; S33111, S33112), or as 400 µL of a 10,000X concentrate in DMSO (S33102). The 1 L unit size provides sufficient material to stain ~20 minigels; the 4 L unit size provides sufficient material to stain ~80 minigels, and is packaged in a cube-shaped container with a removable spigot for easy dispensing and storage.

The SYBR Safe DNA Gel Stain Starter Kit (S33110) includes 1 L of SYBR Safe gel stain in 0.5X TBE buffer and one SYBR Safe photographic filter (S37100).

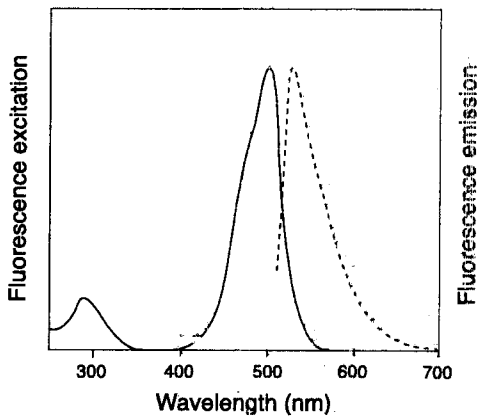

Figure 1. Normalized fluorescence excitation and emission spectra of SYBR Safe DNA gel stain, determined in the presence of DNA.

Storage

SYBR Safe DNA gel stain is stable for at least 6 months when stored at room temperature. The stain is provided in a light-proof container and should not be stored in a secondary container where it could be exposed unnecessarily to light.

Handling and Disposal

SYBR Safe DNA gel stain showed no or very low mutagenic activity when tested by an independent, licensed testing laboratory, and this stain is not classified as hazardous waste under U.S. Federal regulations. The safety testing included three well-established mammalian cell–based tests (Table 1), a battery of well-established Ames-test bacterial strains (Figure 2) and extensive testing for environmental safety (Table 2). Nevertheless, please exercise appropriate care and judgment when using this reagent, and dispose of the stain in compliance with all pertaining local regulations.

Protocols

Staining Nucleic Acids after Electrophoresis

1.1 Soak the gel in SYBR Safe stain. If using SYBR Safe gel stain concentrate, dilute 10,000X in TAE or TBE buffer (as

Table 1. Summary of mammalian cell–based tests for DNA genotoxicity.

Test *	Cell Type	Test Result with S9 Activation †	Test Result without S9 Activation †
Transformation test [1]	Syrian hamster embryo (SHE) cells	Not applicable	Negative
Chromosomal aberration test [2]	Cultured human peripheral blood lymphocytes	Negative	Negative
Forward-mutation test [3,4]	L5178Y TK mouse lymphoma cells	Negative	Negative

* All tests were performed by Covance Laboratories, Inc., Vienna, VA, an independent testing laboratory. † S9, a mammalian extract obtained from Aroclor™ 1254-induced rat liver.

1. Fundamental and Molecular Mechanisms of Mutagenesis 356:1 (1996); **2.** Evans, H.J., in *Chemical Mutagens. Principles and Methods for their Detection Vol 4.* A. Hollaender, Ed., Kluwer Academic/Plenum Publishers (1976) pp. 1–29; **3.** Mutation Res 72, 447 (1980); **4.** Mutation Res 59: 61 (1979).

appropriate) prior to use. Place the gel in a plastic container, such as a pipet-tip box lid or a household food-storage container. Do not use a glass container, as the dye in the staining solution may adsorb to the walls of the container, resulting in poor gel staining. Add sufficient SYBR Safe DNA gel stain to cover the gel. A 50 mL volume is sufficient for staining most standard minigels. To stain larger gels, increase the volume of staining solution in proportion to the increased gel volume, and ensure that the entire gel is fully immersed during staining.

1.2 Incubate for 30 minutes. Protect the gel and staining solution from light by covering it with aluminum foil or by placing it in the dark. Gently and continuously agitate the gel at room temperature (e.g., on an orbital shaker at 50 rpm). No destaining is required.

Precasting SYBR Safe Stain in Agarose Gels

2.1 Prepare the agarose gel directly in SYBR Safe DNA gel stain. SYBR Safe stain is provided in buffer; simply substitute SYBR Safe stain for the buffer when preparing the molten agarose. If using the concentrate, dilute appropriately prior to use. The agarose/SYBR Safe stain mixture may be heated in the microwave. As with precasting gels with ethidium bromide, the mobility of nucleic acid fragments in the gel may be somewhat slower when run in these gels, compared to their mobility in the gel without stain.

2.2 Run the gel. Use a running buffer appropriate to the SYBR Safe gel stain formulation. No post-staining or destaining is needed.

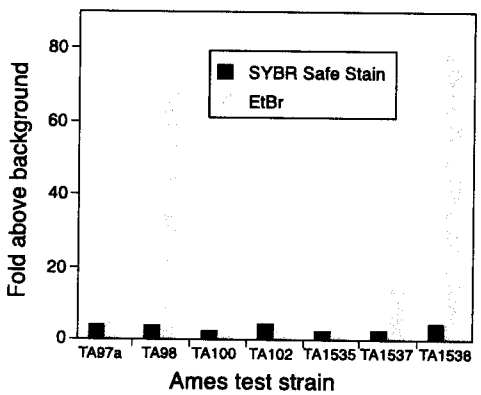

Figure 2. Summary of Ames test results for mutagenicity. Samples were pre-treated with a mammalian S9 fraction and then tested using the indicated Ames test strain. With strains TA97a, TA98, TA100 and TA102, a result of less than twofold above background suggests that the compound is nonmutagenic in the test; whereas, a result of greater than this value suggests that the compound is mutagenic in the test. With strains TA1535, TA1537 and TA1538, a result of less than threefold above background suggests that the compound is nonmutagenic in the test; whereas, a result of greater than this value suggests that the compound is mutagenic in the test. All tests were performed by Covance Laboratories, Inc., Vienna, VA, an independent testing laboratory.

Viewing and Photographing the Gel

Stained gels can be viewed using a standard 300 nm transilluminator, a 254 nm epi- or transilluminator or a blue-light transilluminator such as the Clare Chemical DarkReader™ transilluminator. DNA stained with SYBR Safe stain can also be visualized and analyzed using imaging systems equipped with an excitation source in the UV range or between 470–530 nm. Refer to the excitation/emission characteristics of SYBR Safe stain (Figure 1) in selecting the optimal filter sets to use, or contact the instrument manufacturer for advice.

Stained gels can be photographed using Polaroid® 667 black-and-white print film and SYBR Safe photographic filter (S37100). Molecular Probes' SYPRO® photographic filter (S6656) or a Kodak® Wratten #9 filter will also work well. Using this film and one of these filters, SYBR Safe DNA gel stain provides approximately twice the detection sensitivity as ethidium bromide using a photographic filter appropriate for ethidium bromide. A standard ethidium bromide photographic filter is not appropriate for use with SYBR Safe DNA gel stain. Gels stained with SYBR Safe stain can also be imaged using a CCD camera or a laser-based scanner.

Table 2. Summary of environmental safety test results.

Analysis *	Method	Results
Aquatic toxicity	Fathead minnow CA Title 22 acute screening	Not classified as hazardous or toxic to aquatic life
Ignitability	EPA 1010	Not ignitable (>212°F)
Corrosivity	EPA 150.1	Not corrosive (pH = 8.25)
Corrosivity (by Corrositex®)	DOT-E 10904	Category 2 noncorrosive
Reactivity	EPA 9010B/9030A	No reactivity detected

* All tests were independently confirmed by AMEC Earth and Environmental San Diego Bioassay Laboratory, San Diego, CA.

Product List *Current prices may be obtained from our Web site or from our Customer Service Department.*

Cat #	Product Name	Unit Size
S33100	SYBR Safe™ DNA gel stain in 0.5X TBE	1 L
S33101	SYBR Safe™ DNA gel stain in 0.5X TBE	4 L
S33102	SYBR Safe™ DNA gel stain *10,000X concentrate in DMSO*	400 µL
S33110	SYBR Safe™ DNA Gel Stain Starter Kit *with 1 L of SYBR Safe™ DNA gel stain in 0.5X TBE (S33100) and one photographic filter (S37100)*	1 kit
S33111	SYBR Safe™ DNA gel stain in 1X TAE	1 L
S33112	SYBR Safe™ DNA gel stain in 1X TAE	4 L
S37100	SYBR Safe™ photographic filter	each

Contact Information

Further information on Molecular Probes' products, including product bibliographies, is available from your local distributor or directly from Molecular Probes. Customers in Europe, Africa and the Middle East should contact our office in Leiden, the Netherlands. All others should contact our Technical Assistance Department in Eugene, Oregon.

Please visit our Web site — **www.probes.com** — for the most up-to-date information.

Molecular Probes, Inc.
29851 Willow Creek Road, Eugene, OR 97402
Phone: (541) 465-8300 • Fax: (541) 335-0504

Customer Service: 6:00 am to 4:30 pm (Pacific Time)
Phone: (541) 335-0338 • Fax: (541) 335-0305 • order@probes.com

Toll-Free Ordering for USA and Canada:
Order Phone: (800) 438-2209 • Order Fax: (800) 438-0228

Technical Assistance: 8:00 am to 4:00 pm (Pacific Time)
Phone: (541) 335-0353 • Toll-Free (800) 438-2209
Fax: (541) 335-0238 • tech@probes.com

Molecular Probes Europe BV
Poortgebouw, Rijnsburgerweg 10
2333 AA Leiden, The Netherlands
Phone: +31-71-5233378 • Fax: +31-71-5233419

Customer Service: 9:00 to 16:30 (Central European Time)
Phone: +31-71-5236850 • Fax: +31-71-5233419
eurorder@probes.nl

Technical Assistance: 9:00 to 16:30 (Central European Time)
Phone: +31-71-5233431 • Fax: +31-71-5241883
eurotech@probes.nl

Molecular Probes' products are high-quality reagents and materials intended for research purposes only. These products must be used by, or directly under the supervision of, a technically qualified individual experienced in handling potentially hazardous chemicals. Please read the Material Safety Data Sheet provided for each product; other regulatory considerations may apply.

Molecular Probes' SYBR Safe™ DNA gel stain is licensed under existing patents solely for use in staining nucleic acids in gels.

Several of Molecular Probes' products and product applications are covered by U.S. and foreign patents and patents pending. Our products are not available for resale or other commercial uses without a specific agreement from Molecular Probes, Inc. We welcome inquiries about licensing the use of our dyes, trademarks or technologies. Please submit inquiries by e-mail to busdev@probes.com. All names containing the designation ® are registered with the U.S. Patent and Trademark Office.