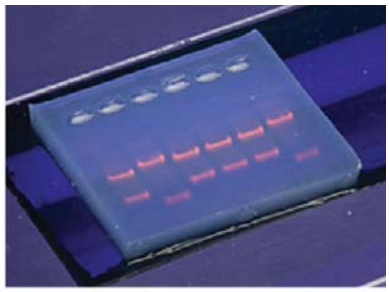
EDVOTEK Quick Guide:

Visualizing DNA

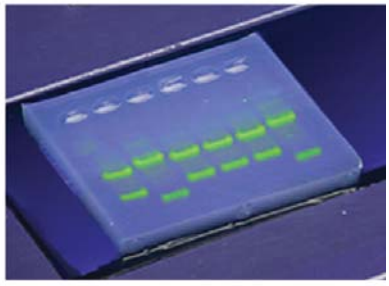
Agarose gel electrophoresis is used to separate DNA fragments in complex mixtures according to their size. However, because DNA is clear and colorless, these bands cannot be seen with the naked eye. Edvotek® offers several different methods for visualizing the DNA separated by electrophoresis.

Fluorescent DNA Stains:

Research laboratories commonly use fluorescent DNA stains because they are extremely sensitive, making it easy to quantify small amounts of DNA. In order to visualize the DNA fragments, an ultraviolet (UV) light source (such as a transilluminator) is used to excite the fluorescent molecules. We offer two fluorescent DNA stains: InstaStain® Ethidium Bromide and SYBR® Safe DNA Stain.



InstaStain® Ethidium Bromide



SYBR® Safe DNA Stain

Visible Dye-based DNA Stains:

Although they are less sensitive than fluorescent stains, dye-based DNA stains are an excellent alternative for the teaching classroom, as they are non-toxic and require no special equipment for visualization. The molecules of the DNA stain possess a positive charge, which allows them to bind to the negatively charged backbone of DNA. The DNA fragments are easily visualized because the bound dye molecules stain them with an intense blue color. We offer two visible dye-based DNA Stains: InstaStain® Blue and Flash Blue Stain.

InstaStain® Blue and FlashBlue™

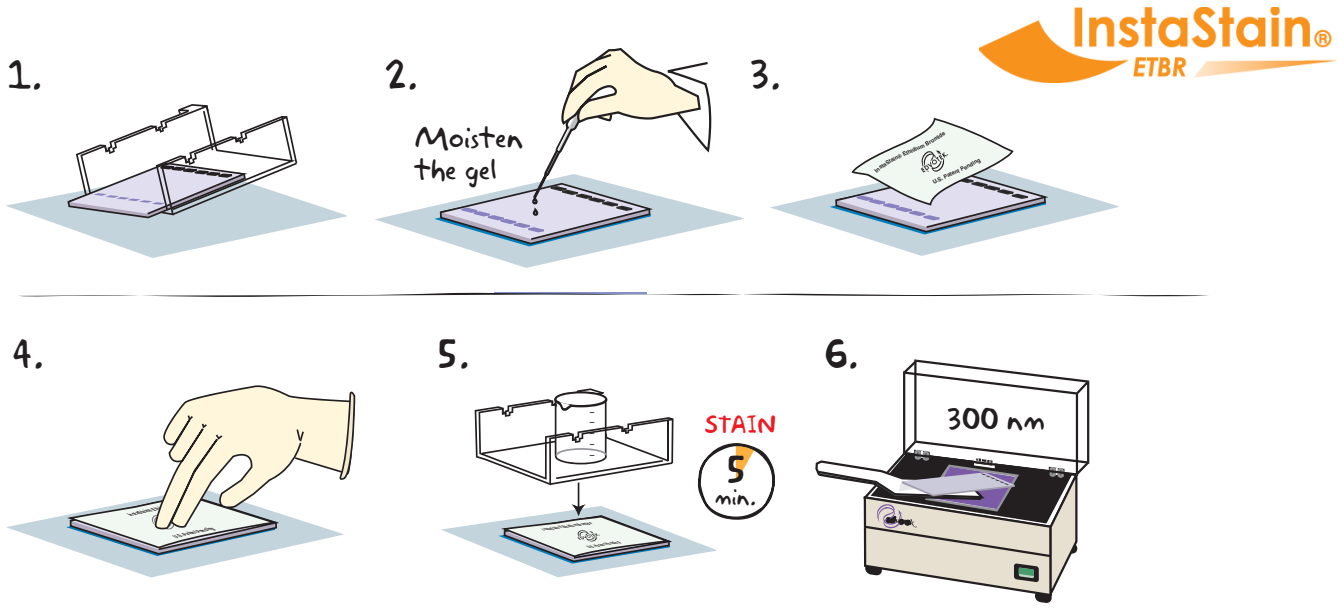
Which DNA Stain Should I Use?

Stain	Advantages	Disadvantages
InstaStain® EtBr	Very sensitive Very fast	Requires UV transilluminator Potentially mutagenic
SYBR® Safe	Very sensitive Non-mutagenic	Requires UV transilluminator More expensive
InstaStain® Blue	Easy to use Generates minimal waste	Less sensitive More time
FlashBlue™	Simple and fast Reusable, inexpensive	Less sensitive Disposal of liquid

EDVOTEK®

InstaStain® Ethidium Bromide

The most commonly used fluorescent DNA stain is Ethidium Bromide (EtBr). Individual EtBr molecules can squeeze between neighboring base pairs in a DNA double helix in a process known as "intercalation". When excited with UV light, any EtBr intercalated into the DNA fluoresces and produces a bright orange light. However, because EtBr is a potential mutagen, it must be handled with care. InstaStain® Ethidium Bromide provides the sensitivity of EtBr while minimizing potential contact with hazardous materials by delivering a small amount of stain to the agarose gel via a special paper backing.



WEAR GLOVES AND GOGGLES WHEN USING THIS PRODUCT.

1. Carefully **REMOVE** the agarose gel and casting tray from the electrophoresis chamber. **SLIDE** the gel off of the casting tray on to a piece of plastic wrap on a flat surface.

   DO NOT STAIN GELS IN THE ELECTROPHORESIS APPARATUS.

2. **MOISTEN** the gel with a few drops of electrophoresis buffer.

3. Wearing gloves, **REMOVE** and **DISCARD** the clear plastic protective sheet from the unprinted side of the InstaStain® card(s). **PLACE** the unprinted side of the InstaStain® Ethidium Bromide card(s) on the gel. Each InstaStain® Ethidium Bromide card will stain 49 cm^2 of gel (7 x 7 cm).

4. With a gloved hand, **REMOVE** air bubbles between the card and the gel by firmly running your fingers over the entire surface. Otherwise, those regions will not stain.

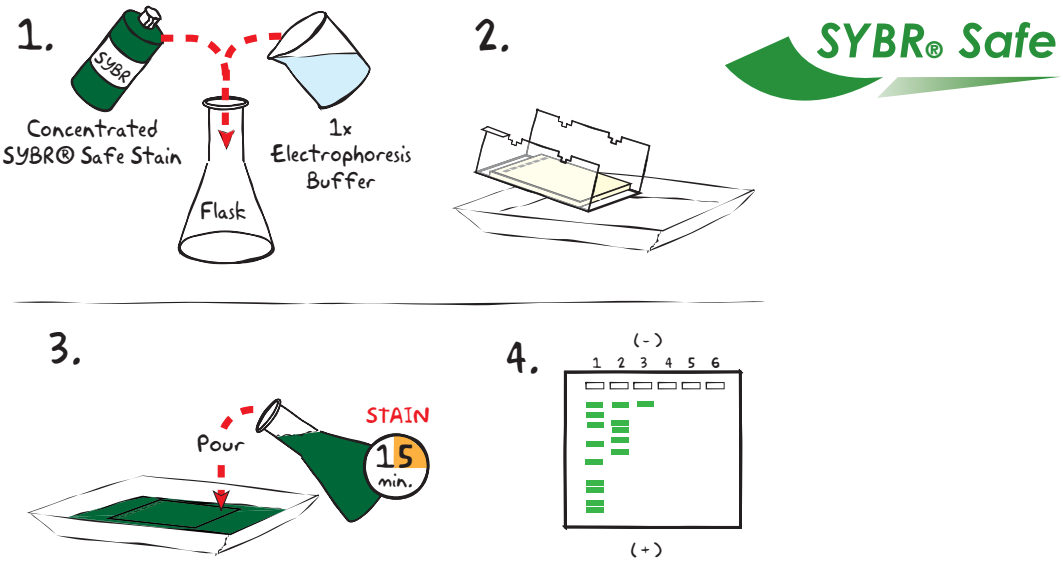
5. **PLACE** the casting tray on top of the gel/card stack. **PLACE** a small weight (i.e. an empty glass beaker) on top of the casting tray. This ensures that the InstaStain® Ethidium Bromide card is in direct contact with the gel surface. **STAIN** the gel for 3-5 min. for an 0.8% gel or 8-10 min. for a gel 1.0% or greater.

6. **REMOVE** the InstaStain® Ethidium Bromide card(s). **VISUALIZE** the gel using a long wavelength ultraviolet transilluminator (300 nm). DNA should appear as bright orange bands on a dark background.

   BE SURE TO WEAR UV-PROTECTIVE EYEWEAR!

SYBR® Safe DNA Stain

SYBR Safe® is a DNA stain that fluoresces with a bright green color when excited with UV light. Similarly to EtBr, SYBR Safe® binds specifically to the DNA double helix. Unlike EtBr, SYBR Safe® has been engineered to be less mutagenic then EtBr, making it much safer to use, particularly in the classroom.



WEAR GLOVES AND GOGGLES WHEN USING THIS PRODUCT.

1. **DILUTE** SYBR® Safe 1: 10,000 by adding 7.5 microliters of the concentrated stain to 75 mL of 1x electrophoresis buffer in a flask. **MIX** well.

2. **REMOVE** the agarose gel and casting tray from the electrophoresis chamber. **SLIDE** the gel off of the casting tray into a small, clean gel-staining tray.

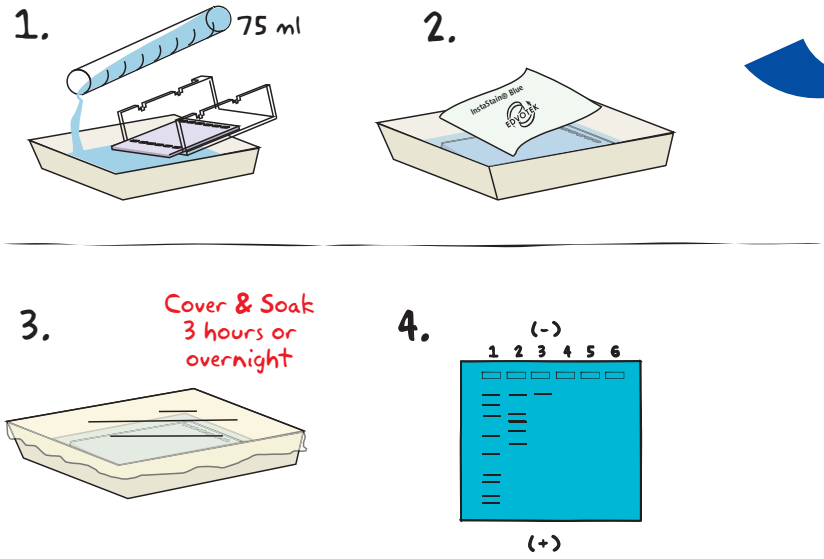
3. **COVER** the gel with the 1x SYBR® Safe stain solution. **COVER** the tray with foil to protect the gel from light. **STAIN** the gel for 10-15 minutes. For best results, use an orbital shaker to gently agitate the gel while staining.

4. **REMOVE** the gel from the staining solution. **VISUALIZE** results using a U.V. transilluminator. DNA bands will appear bright green.

   BE SURE TO WEAR UV-PROTECTIVE EYEWEAR!

InstaStain® Blue

The easiest and most convenient DNA stain available is InstaStain® Blue. InstaStain® Blue does not require the formulation, storage and disposal of large volumes of liquid stain. Each InstaStain® Blue card contains a small amount of blue DNA stain. When the card is placed in water, the DNA stain is released. This solution simultaneously stains and destains the gel, providing uniform gel staining with minimal liquid waste and mess.



WEAR GLOVES AND GOGGLES WHEN USING THIS PRODUCT.

DO NOT STAIN GELS IN THE ELECTROPHORESIS APPARATUS.

1. Carefully **SLIDE** the agarose gel from its casting tray into a small, clean tray containing at least 75 ml of distilled/deionized water or used electrophoresis buffer. The gel should be completely submerged.

   Note: Appropriate staining trays include large weigh boats and small, plastic food containers.

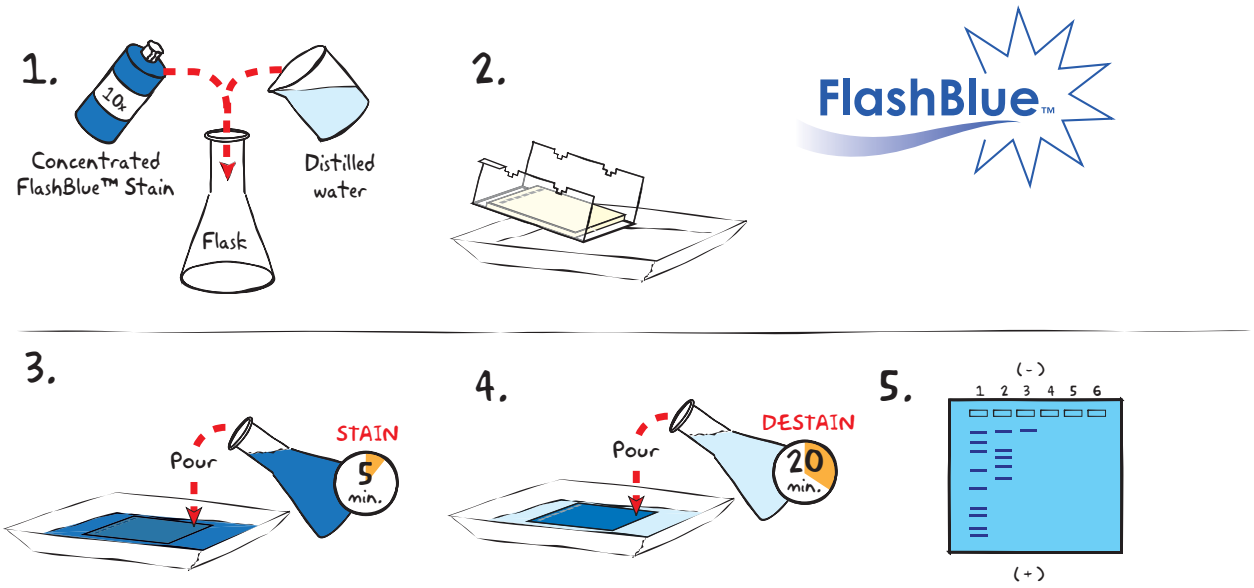
2. Gently **FLOAT** the InstaStain® Blue card(s) on top of the liquid with the stain (blue side) facing the gel. Each InstaStain® Blue card will stain 49 cm^2 of gel (7 x 7 cm). **REMOVE** the InstaStain® card(s) after 30 seconds.

3. **COVER** the tray with plastic wrap to prevent evaporation. **SOAK** the gel in the staining liquid for at least 3 hours. The gel can remain in the liquid overnight if necessary.

4. Carefully **REMOVE** the gel from the staining tray and **DOCUMENT** results.

FlashBlue™ Stain

FlashBlue™ is a proprietary DNA stain that offers simple and rapid staining of agarose gels. FlashBlue™ is provided as a concentrated liquid stain that, when diluted, can be used for both rapid and overnight staining of DNA fragments.



WEAR GLOVES AND GOGGLES WHEN USING THIS PRODUCT.

1. **DILUTE** 10 ml of 10x concentrated FlashBlue™ with 90 mL of water in a flask and **MIX** well.

2. **REMOVE** the agarose gel and casting tray from the electrophoresis chamber. **SLIDE** the gel off of the casting tray into a small, clean gel-staining tray.

3. **COVER** the gel with the 1x FlashBlue™ stain solution. **STAIN** the gel for 5 minutes. For best results, use an orbital shaker to gently agitate the gel while staining. **STAINING THE GEL FOR LONGER THAN 5 MINUTES WILL REQUIRE EXTRA DESTAINING TIME.**

4. **TRANSFER** the gel to a second small tray. **COVER** the gel with water. **DESTAIN** for at least 20 minutes with gentle shaking (longer periods will yield better results). Frequent changes of the water will accelerate destaining.

5. **REMOVE** the gel from the destaining liquid. **VISUALIZE** results using a white light visualization system. DNA will appear as dark blue bands on a light blue background.

Alternate Protocol:

1. **DILUTE** one mL of concentrated FlashBlue™ stain with 149 mL dH_2O.
2. **COVER** the gel with diluted FlashBlue™ stain.
3. **SOAK** the gel in the staining liquid for at least three hours. For best results, stain gels overnight.

Equipment

Cat. # 5062

Classroom DNA Electrophoresis LabStation™

Supports up to 24 Students

Includes:
One M36 HexaGel™ Electrophoresis Apparatus (Cat. #515), one EVT 300 Dual Power Supply (Cat. #509), two 40 µl Fixed Volume MiniPipets (Cat. #588), one order of Yellow Micropipet Tips (1 - 200 µl / 2 Racks of 96) (Cat. #636), and one DNA Fingerprinting Classroom Kit (Cat. #130).

Cat. #558

Midrange UV Transilluminator

7 x 14 cm UV filter

The all-new Midrange UV Transilluminator is compact and designed to visualize DNA stained with Ethidium Bromide, Sybr® Safe, and other fluorescent stains. The UV filter size is 7 x 14 cm and is optimal for visualizing all of our gel sizes. Safety features include a UV-blocking cover and a power cut-off switch when the cover is opened.

InstaStain® Ethidium Bromide

Rapid, sensitive and contains only a few micrograms of ethidium bromide. In 2 minutes, an agarose gel is ready for visualization. Disposal is minimal compared to the volume of liquid waste generated from the standard ethidium bromide staining procedure.

For 40 gels, 7 x 7 cm
Cat. # 2001

For 100 gels, 7 x 7 cm
Cat. # 2002

SYBR® Safe Stain

- SAFE for the Biotechnology Classroom!
- More sensitive than ethidium bromide
- Non-mutagenic

Save time, money, the environment...and get better gel results!

Concentrate-
for 750 ml
Cat. # 608

FlashBlue™ DNA Staining System

FlashBlue™ is a proprietary visible light DNA stain that has been optimized to shorten the time required for both staining and destaining steps.

10X Concetrate, For 3 L
Cat. # 609

InstaStain® Blue

InstaStain® Blue sheets stain gels in minutes and give high quality and uniform gel staining with excellent results for photography. They are environmentally friendly, avoiding large amounts of liquid stain and waste disposal.

For 40 gels, 7 x 7 cm
Cat. # 2003

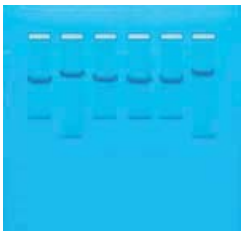
For 100 gels, 7 x 7 cm
Cat. # 2004

Experiments

Cat. # 109

DNA Fingerprinting by Restriction Enzyme Patterns

Basic concepts of DNA fingerprinting are featured in this lab by comparing crime scene DNA with suspect DNAs. Fingerprint patterns are separated by agarose gel electrophoresis and the students determine who may have done-it!



Cat. # 212

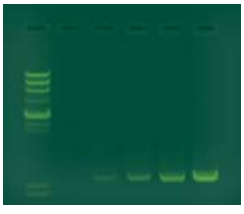
Cleavage of Lambda DNA with *Eco* RI

The DNA from bacteriophage lambda is a well-characterized linear molecule containing six recognition sites for Eco RI (generating 5 fragments with distinct sizes and 2 fragments that are very close in size). In this experiment, Lambda DNA is digested by the Eco RI endonuclease. The digestion products are analyzed by agarose gel electrophoresis.

Cat. # 330

PCR Amplification of DNA

In this easy PCR experiment, students will make billions of copies of a small amount of DNA in just 90 minutes! They will just need to mix template DNA & primers with PCR beads that contain all of the other components required to carry out a PCR reaction. Students will see the increasing amounts of DNA for themselves, taking samples every few cycles and analyzing them on a DNA gel.



Cat. # 332

Mitochondrial DNA Analysis Using PCR

The mitochondria are thought to have evolved from a symbiotic relationship between prokaryotic and eukaryotic cells. Mitochondria have their own DNA and are only inherited via the maternal line. In this experiment, your students will amplify two regions of their mitochondrial DNA.