DNA Biotechnology Kit
NEW
Viewing DNA Gels using SYBR™Safe DNA Stain and the Edvotek TruBlu™2 Transilluminator

Description. We have a new system for visualizing DNA fragments. The DNA Kit now contains **SYBR Safe DNA Stain**, a fluorescent stain that binds specifically to the DNA double helix. When excited with blue light, the stain fluoresces with a green/yellow color. Preparation is easy. Add SYBR Safe directly to the 1X TAE buffer used to prepare the agarose gels, then prepare gels following the standard procedure described in the DNA Kit manual. The kit contains two Edvotek TruBlue2 Transilluminators for viewing the gels. Students can move the gel from the electrophoresis chamber to the transilluminator and see results immediately. For expendable-only loans, we will continue to ship methylene blue when requested.

Preparation of Agarose Gels using SYBR Safe DNA Stain

Reagents

1X TAE buffer prepared from 50X TAE stock
Agarose powder
SYBR Safe DNA Stain, in brown opaque tube (store room temp) ------>

Procedures

Summary: Prepare the 1X TAE buffer as described in the DNA Kit manual (p. 27, repeated below). Before adding the agarose, add SYBR Safe directly to the 1X TAE buffer and swirl to mix. The mixture will be a pale red color, as shown in the photos. Prepare agarose gels according to kit instructions.

1. **Dilute 50X TAE to 1X concentration.** Dilute 1:50 in water (distilled, if available) for the 1X concentration. You will need 30 ml of 1X TAE to prepare each 30 ml agarose gel.

To prepare 1X TAE (amounts include a cushion):

For:	1 gel[1]	4 gels	8 gels (1 class)	16 gels (2 classes)	24 gels (3 classes)	32 gels (4 classes)	….
50X TAE	1 ml	3 ml	6 ml	12 ml	18 ml	24 ml	…
water	49 ml	147 ml	294 ml	588 ml	882 ml	1,176 ml	…

[1] Note: the smallest volume that can be dispensed by the micropipette provided with the kit is 5 µl. For one gel, prepare 50 ml 1X TAE + 5 µl SYBR Safe, but only use 30 ml of the solution to prepare the agarose gel.

2. **Add the SYBR Safe DNA stain as follows** (provided for preparing 1-8 agarose gels). Spin the SYBR Safe vial briefly (5-10 seconds) in the microcentrifuge before using:

For:	1 gel*	2 gels	3 gels	4 gels	5 gels	6 gels	7 gels	8 gels
1X TAE	50 ml*	60 ml	90 ml	120 ml	150 ml	180 ml	210 ml	240 ml
SYBR Safe	5 µl	6 µl	9 µl	12 µl	15 µl	18 µl	21 µl	24 µl

* Prepare 50 ml of the 1X TAE/SYBER Safe solution, but only use 30 ml for one gel.

3. Use the 1X TAE/SYBR Safe solution to prepare the 0.9% agarose gels as described in the manual (p. 28). Run the gels following the procedures on page 29 of the manual.

NOTES

- If the instructor is preparing the agarose solution, but the students will pour the gels, the agarose solution can be made up the day before and stored in a 60°C water bath. Cover the container well. Provide students with 30 ml aliquots. 50-ml capped conical centrifuge tubes work well for aliquoting 30 ml; the tubes can be placed in the water bath until needed.
- The gels can be poured one day and used on another day. After the gels are poured and solidified, place the gel tray in a container that can be sealed (e.g., a large plastic container or sealable plastic bag). Place damp paper towels in the bottom of the container to prevent the gels from drying out.

Viewing an Agarose Gel with the Blue Transilluminator

The blue light of the transilluminator and the orange contrast lid are optimized for SYBR Safe DNA stain. The viewing area can accommodate up to 4 gels at one time.

1. We are currently testing the packaging of the transilluminator. Please remove the unit carefully from the box and the foam liner. Manufacturer instructions are included, but summarized here.
2. Place the unit on a flat surface. For optimum viewing, do not use near a window or where there is high ambient light (it may be helpful to turn off the room lights when photographing the gel).
3. Connect the power cord to the connector at the back of the unit. Plug into an outlet.
4. After the gel has electrophoresed for the desired amount of time, turn off the power supply and disconnect the leads.
5. (Wear gloves) Remove the gel box lid and remove the gel tray from the gel box. Place the tray with gel in a plastic dish. Do not let the gel slide off the gel tray.
6. Transport the tray/gel to the light box, raise the orange lid and slide the gel onto the blue plate of the light box. **Close the orange lid** and turn on the blue light by pressing the front power button up to the top position (black circle).
7. Visualize the DNA bands. Capture an image of the gel to analyze the gel fragments; include the wells in the image (p. 20, DNA Kit manual).
8. Turn off the blue light by moving the switch to the middle position (empty circle). Remove the gel. Wipe the blue plate using a soft tissue and (distilled) water.

PACKING THE TRANSILLUMINATOR FOR RETURN SHIPPING

*****NOTE: We are in the testing phase for shipping the transilluminators, so please do not hesitate to provide feedback to biotechinabox@vt.edu*****

1. Gently wipe the blue plate with a soft cloth; moisten the cloth with water if needed.
2. Remove the power cord and put it in the labeled plastic bag.
3. Place the light box in the original cardboard box with the foam liner. Put the thin foam sheet on top of the orange cover and close the cardboard box. Place the cardboard box in the bubble bag.
4. Stack the 2 transilluminators in the gray crate and pack returned materials/bubble wrap in the crate to prevent the boxes from moving during shipping.