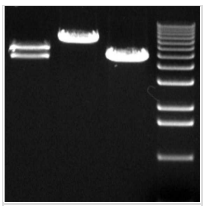

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Agarose Gel Electrophoresis (Instructor Protocol)

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Yeast ORFans CURE

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ABSTRACT

This is the instructor protocol for

Agarose gel electrophoresis
by Brian Teague,
University of Wisconsin - Stout

PREVIEW RUN

The protocol is pretty straightforward, but I have included several common student errors to watch out for!

PROTOCOL CITATION

Brian Teague 2022. Agarose Gel Electrophoresis (Instructor Protocol). **protocols.io** https://protocols.io/view/agarose-gel-electrophoresis-instructor-protocol-ce6nthde

IMAGE ATTRIBUTION

Dr d12, CC BY-SA 3.0 , via Wikimedia Commons

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MATERIALS TEXT

LAB buffer (recipe linked in the steps)

SAFETY WARNINGS

Lithium acetate: May cause eye and skin irritation. May cause respiratory and digestive tract irritation. The toxicological properties of this material have not been fully investigated.

Boric acid: May damage fertility. May damage the unborn child.

Wear appropriate personal protective equipment (PPE), including a lab coat, nitrile gloves and safety glasses.

Make the LAB buffer

1 Make LAB buffer following this recipe:

LAB Agarose Gel Electrophoresis Buffer Recipe
by Brian Teague,
University of Wisconsin - Stout

PREVIEW RUN

For a class of 24, I usually prepare 6x **1 L** bottles of 1X LAB. We go through a lot -- there are three gels that get run over the course of the semester, plus repeats.

Instructor Tips & Common Student Errors

2 **Instructor Tips**

- The benefit of using LAB is the ability to run gels at higher voltage -- our "mini" gel tanks go at 120 V for 20 minutes and get pretty decent gels. It is totally possible to cast, load and run an LAB gel in 2 hours.
- I usually demonstrate the proper assembly of the gel tank
- Gels with Sybr Safe are fine in the refrigerator for 48 hours, but much longer than that and the stain degrades too much. That is -- a gel cast on Tuesday will be fine for a lab on Thursday, but not the other way around. To store a gel, put it in a zipper sandwich baggie and add a splash (not more) of LAB buffer. Label and store at **4 °C** in the refrigerator.
- A common student error is to forget the DNA stain. But all is not lost -- you can post-stain

these gels. Put the gel in a small container (a tip box lid is often fine), cover with LAB, add **5 µL** of Sybr Safe, and put on a slow rocker or orbital shaker.

3 Common Student Errors

- Forgot to add the DNA stain
- Didn't 100% dissolve the agarose (it should be CLEAR AS WATER, no flecks floating around in it)
- Ran the gel backward (remember, "run to red")
- Only ran **1 µL** of the restriction digest (instead of the whole digest)
- Alternately, ran the entire PCR (instead of just **1 µL**) -- leaving not enough to purify.