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Dilutions and Pipetting Practice (Instructor Protocol)

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ABSTRACT

This is the instructor protocol for

Dilutions and Pipetting Practice
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PREVIEW RUN

It contains setup instructions and instructor tips for running the lab.

EXTERNAL LINK

https://qubeshub.org/community/groups/coursesource/publications?id=2556&tab_active=about&v=1

PROTOCOL CITATION

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MATERIALS TEXT

- Grocery store food coloring - red, green, blue, yellow
- (4) 15 ml conicals
- microcentrifuge tubes - (4) for each set.

I usually ask two student groups to share a set of reagents.

SAFETY WARNINGS

Food coloring can stain skin and clothing. Use appropriate PPE, including a lab coat and gloves.

Lab Setup

1 In the conical tubes, add 500 µl of food coloring to 9.5 ml of tap water to make the "10X stocks." Make a stock for each color.

2 Transfer **500 µL** of each stock into a microcentrifuge tubes. Label them appropriately.

3 Follow the student protocol to create standards.

Instructor Tips & Common Student Errors

4 **Instructor Tips**

- I often start the class by having students grab a P-1000 or P-200, setting them to 500 and 200 µl respectively, and together slowly pressing down to find the first stop, then the second. We transfer a bit of water together, then I leave them to their own.
- Students often recognize c1 * v1 = c2 * v2 but have never actually used it (or have forgotten how to use it) to actually make dilutions. The original protocol, from which this was

adapted, has quite explicit instructions. I usually allow students to struggle, ask them to consult their colleagues, etc, and then when a group is properly stuck will help them through it.

- Stand by the standards and compare students' results with them. Students often don't have a good idea of how close is "close enough." Both the volumes and colors should be precisely the same!
- Comparing students' samples to standards is easier if you set the standards on a piece of white paper.
- If a student's sample is substantially different, ask them to go back and try it again.

This is a good place to begin normalizing failure and multiple attempts.

- If, after a second attempt, a student sample isn't correct, follow them back to their bench and observe their technique closely.

It's better to catch technique errors here than later, after a bunch of failed PCRs!

5 Common student errors

- Pressing the plunger all the way down (instead of to the first top) when aspirating, and thus aspirating too much liquid.

This is BY FAR the most common technique error.

- Not watching their pipetting, but doing so blind (especially for the 1/10th scale).
- Making their "mixed" samples from the 10X concentrate instead of the 1X working stocks.
- Forgetting that to make a dilution, they need to add both the concentrate and a diluent (in this case, water).