LAB LESSON PLAN
(Cover Page)

California Agriscience Institute for Agriculture Teachers
California Department of Education

LAB TITLE: PREPARATION OF MEDIA

Ag Model Curriculum Standard(s), Learning Outcomes(s)
& Biological Standard(s)

Addressed: Biological Standards 1, 2, 9, 10, 12, 13, 17

Objective(s): Upon completion of the lab activity, the learner will be able to: Introduce students to the basic concepts of and techniques of tissue culture and micropropagation. To enable students to understand the relationship of plant tissue culture to plant biotechnology. Students appreciate the importance of technological advances in the field of agriculture.

Teacher Preparation: More than one week. Need to order supplies.

How many class periods will lab take? One to three days setup.

Procedures (activities):

Method(s) of Evaluation: Laboratory writeup and teacher observation.
LABORATORY #3 - PREPARATION OF MEDIA

PURPOSE

To learn the basic procedure of preparing media from prepackaged formulations. Nutrient medium contains all the major and minor mineral elements, as well as vitamins, plant growth regulators, sucrose, and a support such as agar. We will be using a commercial powdered preparation in which all mineral, vitamin, and growth regulator components have been weighted for us.

MATERIALS AND METHODS

1. Place a volume of distilled or deionized water, equal to about 2/3 the total volume of media to be prepared, in a beaker.

2. Add contents of packet.

3. Stir until dissolved.

4. Weigh out 30 grams sucrose and add to solution.

5. Adjust pH to 5.7 - 5.8.

6. Bring to total volume of 1000 ml.

7. If you are preparing liquid media, dispense into culture vessels at this point.

8. For agar medium weight, out 5.0 gm Sigma agar. Transfer medium to large flask and add agar. Stir while over head until dissolved. Dispense into culture vessels. Add caps to vessels.

9. Autoclave for 15 min at 121 degrees C. Note: Use flasks or other vessels with a capacity 2 times that of the amount of medium being autoclaved to avoid boiling over.

10. Let medium cool before adding plant material.

DISCUSSION QUESTIONS

1. Why is sucrose added to the medium? Don’t plants get their carbon from photosynthesis?
2. Why are the mineral salts termed 'macronutrients' and 'micronutrients'? (macronutrients include C, H, O, N, P, S, K, Ca, Na, Mg, Cl; micronutrients include I, Bo, Mb, Co, Mn, Cu, Zn, Fe)

3. Are there any other compounds which might be added for plant tissue culture?

**MATERIALS NEEDED FOR LAB**

<table>
<thead>
<tr>
<th>Item</th>
<th>Number Needed/Student or Group</th>
</tr>
</thead>
<tbody>
<tr>
<td>500 or 1000 ml beaker</td>
<td>2</td>
</tr>
<tr>
<td>1000 ml Erlenmeyer flask</td>
<td>1</td>
</tr>
<tr>
<td>Graduated cylinders 1000 ml</td>
<td></td>
</tr>
<tr>
<td>Magnet stirrer</td>
<td>1</td>
</tr>
<tr>
<td>Stirring bar</td>
<td>1</td>
</tr>
<tr>
<td>Package of medium</td>
<td>1</td>
</tr>
</tbody>
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General materials needed for whole class:

- Balance
- Weighing papers
- Sucrose
- Spatulas
- pH meter
- 0.1 and 1.0 N HCL and KOH with droppers
- Agar
- Aluminum foil
- Test tubes and covers
- Spray bottle with isopropyl alcohol for hood
- Marking pens
- Marking tape
- Scissors