

# Laboratory Reagents and Materials

## Equipment

A well-stocked lab will have things such as timers, micropipetters, permanent markers, and ice buckets (as well as ice!). The BioBuilder labs also assume that some consumable materials are available. These consumables are provided if the kits are purchased from Ward's Science, but many labs have the following:

- Nitrile or latex gloves
- 15 ml and/or 50 ml conical screwcap tubes
- Microfuge tubes (1.5–2 ml) and racks
- Sterile test tubes and racks
- Sterile toothpicks, inoculating loops, and/or spreading rods
- 10 cm polystyrene petri dishes—if pouring your own plates

The following specific lab equipment is needed for each of the labs described.

	Roller wheel or shaker	Stir plates and 125 ml Erlenmeyer flasks	Spectrophotometer and cuvettes/glass tubes*	Incubator ** (30°C or 37°C)	Water bath (42°C)
ETS	x	x	x		
iT	x		x		
CW				x	x
GB				x	x

\*Can substitute turbidity standards.

\*\*Can also incubate at room temperature, but requires longer incubation times.

# Reagent Preparation and Storage

Unless otherwise indicated, you can purchase reagents from major chemical vendors, such as Sigma-Aldrich and VWR. They might also be available for lower prices through specialty vendors such as Gold Biotechnology and Ward's Science.

## Solid and Liquid Media

You can also purchase media through vendors such as Teknova.

### *Luria Broth (LB) liquid media*

Dissolve 10 g tryptone, 5 g yeast extract, and 10 g NaCl in water to make 1 L LB media. Autoclave or filter using a 0.2  $\mu$ M filter to sterilize.

### *Luria Broth (LB) agar plates*

Add 10 g tryptone, 5 g yeast extract, 10 g NaCl, and 7.5 g agar to water to make 500 ml LB agar media (enough for ~25 plates). Autoclave to dissolve and sterilize. Pour ~20 ml of LB agar per sterile petri dish. Cool until solidified, then store media side up in the fridge for up to several month. To make LB agar + ampicillin plates, first wait until media is cool enough to touch (~55°C), then add 500  $\mu$ l 1000x ampicillin stock and swirl to mix before pouring into petri dishes as above. Use within one month.

### *Synthetic Complete (SC) minus tryptophan (-Trp) agar plates*

Add 3.4 g yeast nitrogen base without amino acids, 1 g synthetic complete amino acid dropout mix (-Trp), 10 g glucose, and 10 g agar to water to make 500 ml LB agar media. Autoclave to dissolve and sterilize. Pour ~20 ml of SC-Trp agar per sterile petri dish. Cool until solidified, then store media side up in the fridge for up to several months.

### *Yeast extract Peptone Dextrose (YPD) agar plates*

Add 10 g peptone, 5 g yeast extract, 10 g glucose, and 10 g agar to water to make 500 ml YPD agar media. Autoclave to dissolve and sterilize. Pour ~20 ml of YPD agar per sterile petri dish. Cool until solidified, then store media side up in the fridge for up to several months.

## Stock Solutions

### *Ampicillin (100 mg/ml solution)*

Make a 1000x stock by dissolving 100 mg ampicillin in 1 ml sterile water or filter sterilize the solution using a 0.2  $\mu$ M filter. Add 1 ml stock solution to every 1 L of LB or LB+agar for a final working concentration of 100  $\mu$ g/ml. Store frozen stock for up to six months or in the fridge for a week.

*Banana extract (e.g., Frontier Banana Flavor sold at amazon.com)*

Make banana smell standards as described in the Eau That Smell lab protocol. Store standards in fridge for up to several weeks.

*Calcium chloride, CaCl<sub>2</sub> (0.1 M solution)*

Dissolve 735 mg CaCl<sub>2</sub>\*2H<sub>2</sub>O in 50 ml sterile water or filter sterilize the solution using a 0.2 µM filter to make a 0.1 M bacterial transformation solution. Store solution at room temperature.

*Isoamyl alcohol*

Add 700 µl to 1 L LB media. Store LB+isoamyl alcohol in fridge for up to several days.

*Isopropyl β-D-1-thiogalactopyranoside, IPTG (0.1M solution)*

Make a 100x stock by dissolving 24 mg IPTG in 1 ml sterile water or filter sterilize the solution using a 0.2 µM filter. Add 250 µl stock solution to every 25 ml of LB for a final working concentration of 1 mM. Store frozen stock for up to 6 months or in the fridge for a week.

*Ortho-nitrophenyl-β-galactoside, ONPG (0.01 % solution)*

Dissolve 40 mg ONPG in 40 ml water to make a 0.01 % solution. Store frozen for up to several months or in the fridge for a week.

## Buffers

*Bicarbonate buffer (2 % solution)*

Dissolve 1 g sodium bicarbonate or 1 g Arm & Hammer baking soda in 50 ml water to make 2 % bicarbonate solution.

*Carbonate buffer (1M solution)*

Dissolve 5.3 g sodium carbonate or 5.3 gram soda ash (e.g., *Tulip Fashion Art Soda Ash* sold at *amazon.com*) in 50 ml water to make a 1M carbonate solution.

*Lysis buffer (1 % SDS solution or dilute dish soap)*

Dissolve 0.5 g SDS (sodium dodecyl sulfate) or 1 “squirt” clear liquid dish soap in 50 ml water.

## Miscellaneous

*Barium chloride, BaCl<sub>2</sub> (1 % solution)*

Make a 1 % BaCl<sub>2</sub> solution by dissolving 20 mg BaCl<sub>2</sub> in 2 ml water. Approximately 1.75 ml of 1 % BaCl<sub>2</sub> is needed for each complete set of turbidity standards.

*Bleach (10 % solution)*

Add 10 ml of bleach (sodium hypochlorite) to 100 ml water to make 10 % bleach solution.

*Reagents for electronics build in Picture This lab*

See BioBuilder project page on the [Mouser Electronics website](#)

*Sulfuric acid (1 % solution)*

Make a 1 %  $\text{H}_2\text{SO}_4$  solution by adding 1 ml of concentrated  $\text{H}_2\text{SO}_4$  to 99 ml of water. Approximately 80 ml 1 %  $\text{H}_2\text{SO}_4$  is needed for each complete set of turbidity standards. *SAFETY NOTE: Use extreme caution when working with concentrated  $\text{H}_2\text{SO}_4$ . Contact with skin and/or inhalation of fumes should be avoided by working carefully in a hood and using protective personal equipment, such as gloves, lab coat, and safety glasses.*

*Yeast Transformation Kit*

EZ-Yeast™ Transformation Kit by MP Biomedicals #112100200

or

Frozen-EZ Yeast Transformation II Kit by Zymo Research #T2001