FastFred's Media Cookbook (v0.97)(8-30-2004)

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Section A: Basic recipes

Potato Dextrose Agar (PDA)(FDA M127)

---

200 g Potato infusion [dilute to 1L total, ~4g solids]
20 g Dextrose
20 g Agar
1 L Distilled water (dH2O)
To prepare potato infusion, boil 200 g scrubbed, sliced(unpeeled) potatoes in 1 liter distilled water for 30 min. Filter through cheesecloth, saving effluent, which is potato infusion (or use commercial dehydrated form). Mix in other ingredients and boil to dissolve. [Dilute to obtain 1 L final volume] Autoclave 15 min at 121°C. Dispense 20-25 ml portions into sterile 15 x 100 mm petri dishes. Final pH, 5.6 ± 0.2.
Medium should not be re-melted more than once. Medium powder is available commercially but may require supplementing with extra agar to a final concentration of 20 g/liter. To BBL or Difco dehydrated medium, add 5 g of agar.
The broth is clear to slightly opalescent and yellowish in color. [1][2]

Potato Dextrose Yeast (PDY)

---

200 g Potato, infusion from [dilute to 1L total]
20 g Dextrose
2 g Yeast extract
20 g Agar
1 L Distilled water (dH2O)

Gently boil for ten minutes or until the solution is clear.
Autoclave 15 min at 121°C. [13]

Malt Extract Agar (3%)(MEA)(General Microbiology)(FDA M93)

---

30 g Malt extract
20 g Agar
1 L Distilled water
Boil to dissolve ingredients. Avoid overheating, which causes softening of agar and darkening of medium colour. Autoclave 15 min at 121°C. Dispense 20-25 ml into sterile 15 x 100 mm petri dishes. Final pH, 5.5 ± 0.2.
This medium is recommended as a general maintenance medium. [1]

Malt Extract Agar with Yeast (2%)(MEAY)

---

20 g extra light malt extract
2 g yeast
15-20 g agar
1 L water [10]

Sabouraud's Dextrose Broth and Agar (SabDex or SDA)(FDA M133)

---

40 g Dextrose
10 g Polypeptone or neopeptone
1 L Distilled water

Dissolve completely and dispense 40 ml portions into screw-cap bottles. Final pH, 5.8. Autoclave 15 min at 118-121°C. Do not exceed 121°C.
For Sabouraud's dextrose agar, prepare broth as above and add 15-20 g agar, depending on gel strength desired. Final pH, 5.6 ± 0.2. Dispense into tubes for slants and bottles or flasks for pouring plates. Autoclave 15 min at 118-121 °C. [1]
Sabouraud Dextrose (SabDex) Agar is used for the isolation, cultivation, and maintenance of saprophytic and pathogenic yeasts and fungi.
SabDex Agar is an excellent substitute for Malt or Potato Dextrose Agar, when used by mushroom cultivators to propagate mushroom mycelium.
Sabouraud Dextrose Agar was described by Sabouraud in 1892 and was used for the identification of fungi based on their morphological characteristics. Sabouraud Dextrose Agar is a standard medium used to support the growth of yeasts and molds. It supplies peptone as the protein source and dextrose as the carbohydrate source for nourishment. Bacterial suppression occurs due to the low pH. This media is especially suited for the primary isolation of fungi from normally sterile sites such as cerebrospinal fluid (CSF).
Later, Emmons modified the medium by decreasing the dextrose content and adjusting the pH closer to the neutral range. This modification enhances sporulation and is particularly useful for the subculture of fungi that so not develop fruiting structures on other media, and so is useful in their identification. It also serves as a good holding medium for stock cultures. [3]

B. Special Blends

"Karo Water" (Corn Syrup Broth)

---

1 Teaspoon Light Corn Syrup (Karo Syrup or store brand Light Corn Syrup)
100 ml Purified Water
Mix well until dissolved, sterilize for 20-30 minutes at 15 psi. [17]

"Dextrose Tek" Liquid Media (Corn Sugar Broth)

---

1 Teaspoon Powdered Dextrose (corn sugar)
75 ml water [17]

Honey Water (Mycotopia Honey Tek)

---

40 g Honey (or roughly 1 tablespoon per pint of water)
1 L Water
The correct mixture for optimum results is 4% sugars (honey) by weight and 96% water.
Water weighs 1 gram per cc/ml so if you use 100 ml as total weight, then 96 grams/ml/cc of water is mixed with 4 grams of honey, etc.

Sterilize for 30 min at 121°C (15 psi). [14]

Malt-Yeast-Peptone Agar (McKenna's MYP)

---

7 g malt extract (powdered or syrup)
1 g peptone or soytone
0.5 g yeast extract
15 g agar [11]

Malt Yeast Peptone Agar (Stamets MYP)

---

20 g extra light malt extract
1 g yeast extract
1 g peptone
15-20 g agar
1 L water [10]

Grey Cardboard (instead of agar)

Here are the detailed steps for making cardboard plates. Note that you can also use small jars in place of Petri dishes.
1) Measure about 100 mls. of tap water into a small jar.
2) For nutrients, , measure another 100 mls. of tap water into a second jar and add one drop of ordinary soy sauce to the water, and a quarter teaspoon (1.25 mls.) of molasses or light malt powder.
3) Find some gray cardboard, the thicker the better, preferably gray on both sides. Trace a Petri plate onto the cardboard with a pencil and cut out several disks to fit into your plates.
4) Weigh one of your disks and record the weight. Multiply this weight by a factor of 1.3 as a rough guide (you may need to experiment with the amounts here), and add the resulting weight of tap water or nutrient solution to each disk in its Petri plate. (Remember, 1 ounce of water equals 28.35 grams; one gram equals one milliliter.) Example: Suppose my disks weighed 0.17 ounces each. Multiplying 0.17 by 1.3, I get 0.22 ounces. There are 28.35 grams in an ounce, so 0.22 ounces x 28.35 equals 6.3 grams. That means I'll add 6.3 milliliters of solution to each disk.
5) Close up the disks in the plates, and let the water or nutrient solution soak in.
6) Pressure-sterilize the jar of plain water, and the Petri plates with moistened newsprint disks inside, for 10 minutes at 15 psi (allowing the cooker to equilibrate steam for 10 minutes before putting on the pressure regulator).
7) Cool the cooker, and remove the plates and jar of plain water.
8) When the water has cooled, add 3.3 mls. 3% peroxide to the jar, using a pasteurized pipette, to give you a final concentration of about 0.1% peroxide in sterile water.
9) Add about one third of the initial weight of the cardboard as 0.1% peroxide to each disk. Let the solution soak completely into the disks. They are now ready to use. [23]

Oatmeal Flake Agar

---

75 g Oatmeal flakes
20-25 g Agar
1 L water

Stir for 5-10 minutes then filter out the larger particles by pouring it through some mesh, save the broth. [Then add the agar]
This is the best medium for Panaeolus cyanescens I've ever encountered. Beware, this medium will be less firm than the other recipes so extra agar has to be added to compensate. [22]

Moonflower's Rice Malt-Alfalfa-Brewer's Yeast Agar

---

1 cup alfalfa, infusion from
2 cups rice, infusion from
1 standard dolomite (oyster shell-crushed) tablet
1 pkg of regular baker's yeast
Prepare infusion using approx. 1 1/2 quarts of clean water. Mix in the alfalfa and rice, then allow to soak for 2 hours at room temp with occasional stirring. Filter or strain before adding the infusion. Prepare yeast by activating in water for around 30 minutes, then strain out solid yeast grains. Sterilize in pressure cooker for 20 minutes at 15lbs.
This media is reported to produce more "banding" than other media. Will support luxuriant mycelial growth, it is also more than sufficient for starting spores. [19]

MycoPsycho's Liquid Mycelial Culture Broth

1/2 tsp malt (2.5 ml)
3/4 cup water
1. procure a half-pint jar w/ lid & ring and also a spore syringe.
2. drill/punch a hole in the centre of the lid large enough for the syringe.
3. mix 3/4 cup of water with 1/2 tsp.(2.5 ml.) malt.
4. put lid & ring on and put aluminum foil over the top of that and then Pressure Cook (PC) for 20 min. @ 15 psi. , when finished let cool to below 90 degrees F.
5. once cool, setup your sterile environment (flow hood, glove box or even your oven will work) with an alcohol lamp, extra alcohol in a dish, paper towels/napkins, small round bandaid and lastly the spore syringe.
6. remove the foil from the jar, shake the needle and then sterilize the point with the alcohol lamp (get it red hot), cool the needle with a wipe & some alcohol and then inject 1-2 cc/mil spore solution into the jar.
7. once injected wipe the needle w/alcohol and put the guard back on it.
8. cover injection site w/ round band-aid (thanks for that tip magash!).
9. put the jars in your incubator set at 82-84 degrees F. for 4-14 days, you should notice growth by then.
10. sterilize empty syringes in your PC for 15-20 @ 15 psi.
11. remove the bandaid from the jar, sterilize your needle again and then suck myc solution into empty syringe(s) for use on PF style jars or substrate bags. [15]

Ragadinks Liquid Medium

16.5 g dextrose
1.5 g yeast
500 ml tap water [16]

Amaranth Soy Agar

---

20 g amaranth flour
20 g soy flour
9 g agar
500 ml potable or distilled water
Sterilize for 20-30 minutes at 15 psi. [20]

EntheoGenesis No.442

---

10 g amaranth flour
10 g brown rice flour
10 g potato flour
10 g soy flour
2 g malted barley
9 g agar
500 ml potable or distilled water
Sterilize for 20-30 minutes at 15 psi. [20]

Section C: Food Based Recipes and Variations

Corn Meal Agar (CMA)

---

2 g Corn Meal, Infusion from [filter]
15 g Agar
1 L dH2O [4]

Cornmeal Dextrose Agar

---

25 g yellow cornmeal
3 g dextrose
9 g agar
500 ml potable or distilled water [20]

Potato Flake Agar

20.0 g Potato Flakes
10.0 g Dextrose
15.0 g Agar
1.0 L Demineralized Water

Potato Dextrose Agar and Potato Flake Agar are formulations developed to promote sporulation of fungi. The potatoes contained in these media provide nutritious bases for luxuriant growth of fungi. Both media contain dextrose as a growth stimulant.
pH 5.6 +/- 0.2 @ 25°C [7]

Potato Starch Agar

---

30 g potato starch, soluble
20 g dextrose/glucose
15 g agar, pure (omit for liquid media)
1000 mL (d)H2O
The pH is adjusted following autoclaving to prevent agar hydrolysis by acid. [20]

Potato Flake Agar with Yeast

---

20 g potato flakes
10 g glucose
1-2 g dried yeast
20 g agar
1 L water [18]

Potato-Carrot Agar

---

grated potato 20.0 g
grated carrot 20.0 g
Agar 20.0g
Tap water 1000.0 ml
Boil potato and carrot in 1000.0 ml of water for 1 h, filter, add water to the initial volume, adjust pH to 7.0 - 7.1 and add agar.
Sterilize at 121°C for 30 min. [6]

Barley Flour Malt Extract Agar

---

40 g barley flour
2 g malt extract
1 g yeast extract (optional)
9 g agar
500 ml potable or distilled water
Sterilize for 20-30 minutes at 15 psi. [21]

Barley Flour Modified Sabouraud's

---

25 g barley flour
5 g dextrose
2 g Polypeptone or neopeptone (optional)
1 g yeast extract
9 g agar
500 ml potable or distilled water
Sterilize for 20-30 minutes at 15 psi. [21]

Oatmeal Agar (OA)

60.0 g Oatmeal [filter]

12.5 g Agar [may require more]
1.0 L dH2O
Cook oatmeal 5-10 minutes then filter the liquid into another container using cheesecloth or a metal strainer with a tight mesh. Dilute liquid to 1L, add agar, and heat with swirling until solids dissolve. This is reported to be a good media for cultivating Panaeolus cyanescens. [5]

Oatmeal Agar A

Oatmeal 20.0g
Agar 20.0g
Tap water 1000.0 ml
pH 7.2. [6]

Oatmeal Agar [B]

Oats 30.0g
Agar 15.0g
Tap water 1000.0 ml
Keep oats on a water bath at 58°C for 1 h, filter through 2 layers of gauze, dilute to 1000.0 ml and add 15.0 g agar. [6]

V-8 Oatmeal Agar

---
50 ml V-8 juice
25 g Cream of oats
20 g Agar
1 L Water
Be careful to use a container much larger than the volume of medium, i.e., prepare a 500 ml medium in 2 litre flasks or it will tend to boil over no matter how slowly it is cooled down. [20]

V8 Medium

---
50 ml V8 Juice
.2 g $CaCO_3$
20 g of agar
1 L Water
The commercial V8 juice is occasionally used for tissue cultures of edible mushrooms. It should be noted that most mushrooms prefer neutral to slightly acid range of medium, that is, a pH of about 5.5 to 6.5. However the straw mushroom, Volvariella volvacea, prefers a high pH medium, 6.8 to 7.8. Therefore, it is important to make sure the acidity or pH of the medium is correct for a particular mushroom. Here be careful to use a container much larger then the volume of medium, i.e., prepare a 500 ml medium in 2 liter flasks or it will tend to boil over no matter how slowly it is cooled down. [20]

Bean Agar

Beans (peas or pulse) 100.0 g [infusion from (filter)]
K_2HPO_2 0.5g

Sucrose 10.0g
Agar 20.0g
Water 1000.0ml
Prepare infusion from beans. Sterilize at 121°C for 30 min. [6]

Faba Bean Dextrose Agar (FDA)

---

200 g faba bean seeds or 400 g of faba bean leaves [infusion from]
[autoclave and filter to obtain faba infusion]
20 g of dextrose
18 g agar
1 L water
Method:
a. Weigh out 200 g of faba bean seeds or 400 g of faba bean leaves in a 1.5 1 flask. Add 1L of water, and autoclave at 15 psi. for 30 minutes.
b. Pass the autoclaved beans through a sieve, add 18 g of agar, heat, and stir till dissolved.
c. Add 20 g of dextrose, stir till dissolved, and make up the volume to 11 with tap water. d. Autoclave at 15 psi. for 20 minutes, cool to about 40°C, and pour into petri dishes (normally 40 petri dishes/1). This medium is used for propagation of B. fabae, A. fabae, and A. tenuis. [12]

Pea Agar

---

100 g Yellow peas
0.5 g K_2HPO_2
10.0 g Sucrose
20.0 g Agar
1.0 L Tap water

Boil peas in 1000.0 ml of water, filter through gauze, add water to the
initial volume; add phosphate, sucrose, and agar. Sterilize at 121°C for 30 min. [6]

Cabbage Agar

Cabbage 50.0g
glucose 20.0g
Peptone 10.0g
Agar 20.0g
Tap water 1000.0 ml
Boil 50.0 g of cabbage in 1000.0 ml of water, filter cabbage, adjust the
volume of broth to the initial value. [6]

Dr. Pollock's Modified [Dog Food] Agar

---

10 g dried dog food (ground to flour)
10 g amaranth flour
2 g dextrose or malt extract
9 g agar
500 ml potable or distilled water

Sterilize for 20-30 minutes at 15 psi. [20]

Section D: Other Media and Alternate Formulations

Potato-Glucose Agar 1

---

grated potato 200.0 g
glucose 20.0g
Agar 20.0g
Tap water 1000.0 ml
Boil potatoes for 1 h in 1 L of water, filter through gauze. add water to the initial volume, add glucose and agar.
Sterilize at 105°C for 30 min. [6]

Potato-Peptone Medium

Potato decoction 200 ml [infusion from 200g potato]
Yeast extract 1.0 g
Peptone 5.0g
Agar 30.0g
Distilled water 800.0 ml [6]

Potato-Peptone-Yeast Agar (PPYA)

Potato decoction 200 ml [infusion from 200g potato]
200.0 ml
Peptone 5.0g
Yeast extract 1.0 g
Agar 25.0g
Distilled water to 1000.0 ml
pH 8.0. [6]

Malt Agar (MA)(FDA M185)[aka 2% MEA]

---

20 g Malt extract, powdered
20 g Agar
1 L Distilled water
Mix ingredients, steam to dissolve agar and sterilize for 15 min at 121°C. Temper medium to 45°C and pour plates under aseptic conditions.
This medium is recommended as a general maintenance medium. [1]

Difco Malt Extract Broth (FDA M94)

6.0 g Malt extract base
1.8 g Maltose, technical
6.0 g Dextrose

1.2 g Yeast extract
1.0 L Water
Final pH, 4.7 ± 0.2. [1]

Malt Extract Agar for Yeasts and Molds (MEAYM)(FDA M182)

20.0 g Malt extract, powdered
20.0 g Glucose
1.0 g Peptone
20.0 g Agar
1.0 L Distilled water

Mix ingredients, heat to dissolve agar and sterilize at 121°C for 15 min. Temper media to 45°C and pour plates under aseptic conditions. Dehydrated MA is commercially available, but since several MA formulas exist, check for the correct composition.
Final pH 5.4. [1]

Malt Extract Peptone Agar

---

30 g Malt extract
3 g Soya peptone
15 g Agar
1 L Distilled water
Adjust pH to 5.6. Sterilize at 121°C for 10 min. [8]

Raper & Thom MEA (RTMEA)

To 2% MEA add:
Glucose 10g
Soy Peptone 5g [9]

ISP 2 Medium (Malt, Yeast, Glucose)

Malt extract 10.0 g
Yeast extract 4.0 g
Glucose 4.0g
Agar 15.0g
Distilled water 1000.0 ml
pH 7.2. [6]

Yeast Extract Agar (YEA)(FDA M181)

10.0 g Proteose peptone
3.0 g Yeast extract
5.0 g NaCl
15.0 g Agar
1.0 L Distilled water
Adjust pH to 7.2-7.4. Autoclave at 121°C for 15 min. [1]

Yeast Glucose Agar

Yeast extract 5.0 g
Glucose 10.0g
Peptone 5.0g
Agar 20.0g
Distilled water 1000.0 ml
pH 7.2. Sterilize at 121°C for 15 min. [6]

Glucose and Yeast Extract Agar

Glucose 20.0g
Yeast extract 10.0 g
CaCO2 20.0g
Agar 17.0g
Distilled water 1000.0 ml [6]

Glycerin Yeast Agar

Yeast extract 5.0 g
Glycerin 50.0g (also called glycerine or glycerol)
CaCO2 1.0g
Agar 20.0g
Distilled water 1000.0 ml [6]

Manure Tincture

Cow manure (fresh) 1.0 kg
Distilled water 3000.0 ml
Boil, squeeze through gauze into a bottle and dilute 3 to l. [6]

Manure Agar

Horse manure 100-125 g
Agar 25.0g
Distilled water 1000.0 ml
Boil manure in 1000.0 ml of water for 10 min, then keep for 16-20 h, filter through 1-2 layers of filter paper, adjust to the initial volume, add agar. Sterilize at 121°C for 15 min. [6]

Water Agar (aka Starved Agar)

Agar 20.0g
Distilled water 1000.0 ml
Sterilize at 121°C for 15 min. [6]

Gelatin Agar (GA)(FDA M54)

---

4 g Peptone
1 g Yeast extract
15 g Gelatin
15 g Agar
1 L Distilled water
Suspend ingredients with constant stirring to prevent scorching gelatin, and boil to dissolve gelatin and agar. Adjust to pH 7.2 ± 0.2. Autoclave 15 min at 121°C. Cool to 45-50°C. Pour plates. [1]

Plate Count Agar (SMA)(aka Standard Methods Agar)(FDA M124)

5.0 g Tryptone
2.5 g Yeast extract
1.0 g Dextrose
15.0 g Agar
1.0 L Distilled water
Heat to dissolve ingredients. Dispense into suitable tubes or flasks. Autoclave 15 min at 121°C. Final pH, 7.0 ± 0.2.
For viable yeasts and molds, dispense 20-25 ml portions into sterile 15 x 100 mm petri dishes. [1]

Nutrient Agar (FDA M112)

---

3 g Beef extract
5 g Peptone
15 g Agar
1 L Distilled water
Heat to boiling to dissolve ingredients. Dispense into tubes or flasks. Autoclave 15 min at 121°C. Final pH, 6.8 ± 0.2. [1]

Starch Agar (FDA M143)(Nutrient agar with starch)

---

23 g Nutrient agar (FDA M112)
10 g Potato starch
1 L Distilled water

Heat to dissolve agar in 500 ml water. Dissolve starch in 250 ml water. Combine and dilute to 1 liter. Autoclave 15 min at 121°C.
Note: add 3 g agar to Difco's dehydrated starch agar. [1]

Starch-Yeast Agar

Yeast extract 2.0 g
Starch (soluble) 10.0 g
Agar 20.0g
Tap water 1000.0 ml
pH 7.3. [6]

1/5 Starch-Yeast Agar

Yeast extract 0.4 g
Soluble starch 2.0 g
Agar 20.0 g
Distilled water 1000.0 ml
pH 7.3. [6]

Long-term Preservation Medium (FDA M85)

---

3 g Yeast extract, 0.3%
10 g Peptone
30 g NaCl
3 g Agar
1 L Distilled water
Heat to dissolve ingredients. Dispense 4 ml portions to 13 x 100 mm screw-cap tubes. Autoclave 15 min at 121°C. Cool and tighten caps for storage. No pH adjustment is necessary. [1]

Peptone Meat Agar (Meat Water)

Peptone 10.0g
NaCl 5.0g [optional]
Agar 20.0g
Meat water 1000.0 ml
Preparation of meat water: comminute 500 g of meat free of bones, fat and tendons, add 1000.0 ml of tap water and leave for 12 h at room temperature or in a thermostat at 30°C, or for 2 h at 37°C. Then squeeze the meat through gauze or cloth and boil the filtrate for 5 min. The proteins are denatured. Filter the cooled down mass through a cotton-wool filter and add water to the initial volume. pH 7.2 - 7.4. Sterilize at 121°C for 30 min. [6]

E. Common Solutions

Gentamicin Sulfate Solution (FDA M57)

5.00 g Gentamicin sulfate
1.00 L Distilled water
Sterilize by filtration through 0.20 µm membrane. Store at -20°C. [1]

F. Sources

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[16] Ragadinks (Shroomery)
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