

## Common Worm Media and Buffers

Fanglian He

**[Abstract]** Here are recipes of some media and solutions often used in *C. elegans* research.

### **Materials and Reagents**

1. Agar, peptone (BD Biosciences)
2. Cholesterol (Sigma-Aldrich)
3. Streptomycin (Sigma-Aldrich)
4. Nystatin (Life Technologies, Gibco®)
5. Bleach (Clorox)
6. Potassium phosphate
7. Clorox bleach
8. NaCl
9. CaCl<sub>2</sub>
10. MgSO<sub>4</sub>
11. EtOH
12. FeSO<sub>4</sub>·7H<sub>2</sub>O
13. Na<sub>2</sub>EDTA
14. MnCl<sub>2</sub>·4H<sub>2</sub>O
15. ZnSO<sub>4</sub>·7H<sub>2</sub>O
16. CuSO<sub>4</sub>·5H<sub>2</sub>O
17. KH<sub>2</sub>PO<sub>4</sub>
18. Na<sub>2</sub>HPO<sub>4</sub>

### **Equipment**

1. 60 x 15 mm plate
2. Plastic boxes

## Recipes

1. Nematode growth medium (NGM) agar: For the maintenance of worms.

For 1 liter medium

3 g NaCl

17 g agar

2.5 g peptone

1 ml cholesterol (5 mg ml<sup>-1</sup> in 95% EtOH)

975 ml H<sub>2</sub>O

Autoclave, and then add the following sterile solution (autoclaved)

1 ml 1 M CaCl<sub>2</sub>

1 ml 1 M MgSO<sub>4</sub>

25 ml 1 M potassium phosphate (pH 6) (to avoid precipitation, mix between addition of MgSO<sub>4</sub> and potassium phosphate)

To make 1 M potassium phosphate (pH 6): For 1 liter, dissolve 136.1 g KH<sub>2</sub>PO<sub>4</sub> in about 800 ml dH<sub>2</sub>O, then adjust to pH 6.0 with solid KOH (approx 15 g) before bringing up to volume. Make 100 ml aliquots and autoclave.

Need to add streptomycin (300 ng ml<sup>-1</sup>) if plate is used for seeding bacterial food *E coli* OP50-1. Typically pour 60 x 15 mm plate and store NGM plates in plastic boxes with covers at room temperature.

2. S-basal medium (adapted from the Kim Lab at Stanford) : For liquid culture of worms.

For 1 liter medium

5.8 g NaCl

50 ml 25 ml 1 M potassium phosphate (pH 6)

1 ml cholesterol (5 mg ml<sup>-1</sup> in 95% EtOH)

950 ml dH<sub>2</sub>O

Autoclave, and then add the following sterile solution (autoclaved)

3 ml 1 M CaCl<sub>2</sub>

3 ml 1 M MgSO<sub>4</sub>

10 ml trace metals solution

10 ml 1 M potassium citrate (pH 6.0)

10 ml 100x Nystatin (antifungal agent, keep in freezer; do not have to add it all the time).

To make 500 ml trace metals solution

0.346 g FeSO<sub>4</sub>.7H<sub>2</sub>O

0.930 g Na<sub>2</sub>EDTA

0.098 g MnCl<sub>2</sub>.4H<sub>2</sub>O

0.144 g ZnSO<sub>4</sub>.7H<sub>2</sub>O

- 0.012 g  $\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$   
 Sterilize by autoclaving. Keep in dark (wrap in foil).  
 To make 100 ml of 1 M potassium citrate: dissolve 21.02 g citric acid, monohydrate in 80 ml and adjust to pH 6.0 with solid KOH (approx 17g) before bringing up to volume.
3. Worm M9 buffer  
 3 g  $\text{KH}_2\text{PO}_4$   
 6 g  $\text{Na}_2\text{HPO}_4$   
 5 g NaCl  
 Add  $\text{H}_2\text{O}$  to 1 liter. Sterilize by autoclaving.  
 After solution cools down, add 1 ml autoclaved/sterile 1 M  $\text{MgSO}_4$ .
4. 100 ml 2x worm lysis solution: For worm egg prep  
 50 ml dd $\text{H}_2\text{O}$   
 10 ml 10 M NaOH  
 40 ml Clorox bleach  
 Make fresh and store at 4 °C up to one week.