Medium preparation

2022-11-23

Contents

10X M9 salt solution, 1 L

Recipe

Stock	Amount (g)	Concentration (g/L)
Na_2HPO_4 $2H_2O$	85 g	68 g/L
KH_2PO_4	30 g	30 g/L
NaCl	5 g	5 g/L
NH_4Cl	10 g	10 g/L
ddH_2O	1 L	

Element

Stock	Amount (g)	Molar mass	Ratio	Concentration (g/L)	Concentration (mM)
Na_2HPO_4 $2H_2O$	85	142	0.798	68	478.8732
KH_2PO_4	30	136	1.000	30	220.5882
NaCl	5	58	1.000	5	86.2069
NH_4Cl	10	54	1.000	10	185.1852

0.2% glucose with M9 (M9G), 200 mL

Recipe

Stock	Amount	Concentration
glucose powder	0.4 g	0.2% glucose
10X M9	20 mL	1X M9
ddH_2O	180 mL	ddH_2O
1M $MgSO_4$	400 μL	2 mM $MgSO_4$
1M $CaCl_2$	20 μL	0.1mM $CaCl_2$

Item

Item	Usage	Amount
weighing scale, spoon straw and weigh boat	weighing	1
electronic pipette, P1000, P20	dispense	1
50 mL tip	dispense 10X M9	1
250 mL bottle	mixing bottle and filtered bottle	2
200 mL cylinder	dispense	1
stericup	filter	1

Protocol

- **Weigh glucose powder.**
  - Take two 250 mL Pyrex bottles and use tape to label them with `M9G` (mixing bottle) and `M9G filtered` (filtered bottle). Yellow tape is preferred for filtered bottle.
  - Use a spoon straw to scoop glucose powder to the weigh boat in weight scale. Weigh **0.4 g** of glucose powder and pour it in mixing bottle. Throw away the weigh boat and spoon straw and turn off weighting scale. Move the bottles to bench.
- **Set up bench.** Turn on flame, wipe the bench and pipette with 70% ethanol.
- **Add materials.**
  - Use a 50 mL stripette to dispense **20 mL** of 10X M9 to mixing bottle.
  - Use a 200 mL cylinder to dispense **180 mL** of ddH_2O to mixing bottle.
  - Use P1000 set at 400 uL to dispense **400 uL** of $MgSO_4$ to mixing bottle.
  - Use P20 set at 20 uL to dispense **20 uL** of $CaCl_2$ to mixing bottle.
  - Shake the mixing bottle by hand, make sure every solid material is dissolved. If the material is not fully dissolved, use microwave to heat for 30 seconds. Loose the cap before place it in the microwave.
- **Filter media.** on the filtered bottles.
  - Connect the stericup with vacuum pipe. Pour M9G from to mixing bottle into stericup. One hand holds the junction of pipe and stericup, another hand turn on the vacuum. Rotate the faucet until media go through the filter.
  - After the medium has been filtered, disconnect the pipe from filtering machine.
  - Be close to flame while removing the stericup from filtered bottle. Close the cap of filtered bottle.
- **Turn off flame**.

0.2% glucose with M9 (M9G), any volume

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## Total volume: 50 mL
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Stock	Amount	Concentration
glucose	0.1g	0.2%
10X M9	5mL	1X
ddH_20	45mL	
1M $MgSO_4$	100uL	2mM
1M $CaCl_2$	5uL	0.1mM

10X M9 salts, any volume

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## Total volume: 1000 mL
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Stock	Amount	Concentration (g/L)	Concentration (mM)
Na_2HPO_4 $2H_2O$	85g	68	478.87
KH_2PO_4	30g	30	220.59
NaCl	5g	5	86.21
NH_4Cl	10g	10	185.19

10X 0-N Fertilizer