

The Liu Laboratory protocol — BG-11 & MBG-11
Arts & Sci Washington University in St. Louis

BG11 media

10ml 100X BG11 concentrate
1ml 1000X Ferric Ammonium Citrate
1ml 1000X Na₂CO₃
1ml 1000X K₂HPO₄
10ml 1M TES pH 8.2
ddH₂O to 1L

After Autoclaving add 1mL 1000X Vitamins

MBG11 media

Same as BG 11 media but also add:
35g Instant Ocean (dissolve first) (this is for 3.5% IO media for type strain)
1000X Fe-EDTA

BG11 Plates

Autoclave in 500 ml of water in a flask (that is easy to pour from, cap in a way that it can be recapped easily and in a sterile way):

15g Bacto Agar

A stir bar

Autoclave separately these (in a bottle is fine, can remove stir bar):

10ml 100X BG11

1ml 1000X Ferric Ammonium Citrate

1ml 1000X Na₂CO₃

1ml 1000X K₂HPO₄

10ml 1M TES pH 8.2

3g Na thiosulfate

(0.9g glucose for 5mM glucose)

Up to 500ml water

Autoclave 30min liquid cycle, Cool media on the benchtop, agar in the 55°C water bath for 15-30 minutes. Add 1ml 10mg/ml Km for Kanamycin 10µg/ml plates and 2ml for Kan 20 plates to the media, swirl. Remove agar from bath and spray down with ethanol, wipe. Pour media into agar, stir. Pour immediately.

For MBG-11 3.5% IO

Also add to the media:

35g Instant Ocean

1ml 1000X Fe-EDTA

100X BG-11(BG11 concentrate)

For 1L:

149.6g	NaNO ₃
7.5g	MgSO ₄ •7H ₂ O
3.6g	CaCl ₂ •2H ₂ O
0.60g	Citric Acid(or 0.89g Na-citrate, dehydrate)
1.12ml	0.25M Na-EDTA pH 8.0
100ml	Trace Minerals

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Store at 4°C

Trace Minerals

For 1L:

2.68g H₃BO₃

1.81g MnCl₂•4H₂O

0.22g ZnSO₄•7H₂O

0.39g Na₂MoO₄•2H₂O

0.079g CuSO₄•5H₂O

0.049g Co(NO₃)₂•6H₂O

Store at 4°C

1000X Ferric Ammonium Citrate

For 100ml:

600mg Ferric Ammonium Citrate

Store at 4°C shielded from light. Check for growth before use.

1000X Na₂CO₃

For 100ml:

2g Na₂CO₃

Store at 4°C

1000X K₂HPO₄

For 100ml:

3.05g K₂HPO₄

Store at 4°C

1M TES pH 8.2

For 500ml:

114.625g TES

NaOH

Adjust pH to 8.2 with NaOH pellets. Store at 4°C

1000X Fe-EDTA

For 1L:

5.2g Fe-EDTA *This is the medium iron version. **PLEASE USE THIS ONE FOR R61.**

10.4g Fe-EDTA *This is for the high iron version

Store at 4°C

0.25M Na-EDTA pH 8.0

For 1 L:

93.06g Na-EDTA

Adjust pH to 8.0. Store at 4°C