

Supplementary text 1: Composition of the main culture media used in the MNHN collection of cyanobacteria and eukaryotic microalgae.

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Z8 medium (Z8) (Rippka 1988)

Freshwater algae and cyanobacteria

Preparation of stock solutions: **Solution A, B, C and Trace element**

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration mM
A	NaNO ₃	46.7	5.49
	Ca(NO ₃) ₂ , 4H ₂ O	5.9	0.25
	or Ca(NO ₃) ₂	4.1	0.25
	MgSO ₄ , 7H ₂ O	2.5	0.10
B	K ₂ HPO ₄	3.1	0.18
	or K ₂ HPO ₄ , 3H ₂ O	4.1	0.18
	Na ₂ CO ₃	2.1	0.20

For stock solutions **A** and **B**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component	Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1 Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2 (NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3 KBr	1,21	1.02 µM
	T4 KI	0,83	0.50 µM
	T5 ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6 Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7 Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8 CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9 (NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10 Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11 Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O	4,74	0.50 µM
	T12 V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13 H ₃ BO ₃	31 g for 1L	0.05 mM
	MnSO ₄ , 4H ₂ O	22,3 g for 1L	0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with mili-Q water
- Adjust **T12** to **T13** to 1000 mL with mili-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8 medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	10
B	10
C	10
Trace element solution	1

- Adjust to 1000 mL with mili-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Notes:

- For solid culture media :
 - prepare 250 mL of Z8 medium with 5 mL of solution A, B and C and 0,5mL of Trace element solution.
 - Prepare 250 mL of mili-Q water with 6 - 7 g/L of agar in a 500 mL bottle
 - Autoclave at 121°C for 15-20 minutes
 - Mix the 2 solutions under laminar flow hood
 - Place the medium in petri dishes and wait 24 hours before use.
- For semi - solid culture media, add 5 g/L of agar

Z8X medium (Z8X) (Rippka 1988)

Freshwater algae and cyanobacteria

Preparation of stock solutions: **Solution A, B, C and Trace element**

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration mM
A	MgSO ₄ , 7H ₂ O	2.5	0.10
	CaCl ₂ , 2H ₂ O	3.7	0.025
B	K ₂ HPO ₄	3.1	0.18
	or K ₂ HPO ₄ , 3H ₂ O	4.1	0.18
	Na ₂ CO ₃	2.1	0.20

For stock solutions **A** and **B**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component	Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1 Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2 (NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3 KBr	1,21	1.02 µM
	T4 KI	0,83	0.50 µM
	T5 ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6 Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7 Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8 CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9 (NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10 Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11 Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O	4,74	0.50 µM
	T12 V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13 H ₃ BO ₃ MnSO ₄ , 4H ₂ O	31 g for 1L 22,3 g for 1L	0.05 mM 0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with mili-Q water
- Adjust **T12** to **T13** to 1000 mL with mili-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8X medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	10
B	10
C	10
Trace element solution	1

- Adjust to 1000 mL with mili-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Notes:

- For solid culture media :
 - prepare 250 mL of Z8X medium with 5 mL of solution A, B and C and 0,5mL of Trace element solution.
 - Prepare 250 mL of mili-Q water with 6 - 7 g/L of agar in a 500 mL bottle
 - Autoclave at 121°C for 15-20 minutes
 - Mix the 2 solutions under laminar flow hood
 - Place the medium in petri dishes and wait 24 hours before use.
- For semi - solid culture media, add 5 g/L of agar

Z8 Salt medium (Z8 Salt) (Rippka 1988)

Seawater cyanobacteria

Preparation of stock solutions: [Solution A, B, C, Salt Solution and Trace element](#)

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration mM
A	NaNO ₃	46.7	5.49
	Ca(NO ₃) ₂ , 4H ₂ O or Ca(NO ₃) ₂	5.9 4.1	0.25 0.25
	MgSO ₄ , 7H ₂ O	2.5	0.10
B	K ₂ HPO ₄ or K ₂ HPO ₄ , 3H ₂ O	3.1 4.1	0.18 0.18
	Na ₂ CO ₃	2.1	0.20
	Salt Solution	NaCl	87.5
MgSO ₄ , 7H ₂ O		37.5	57

For stock solutions **A**, **B** and **Salt Solution**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component	Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1 Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2 (NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3 KBr	1,21	1.02 µM
	T4 KI	0,83	0.50 µM
	T5 ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6 Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7 Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8 CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9 (NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10 Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11 Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O (ou KAl(SO ₄) ₂ , 12H ₂ O)	4,74	0.50 µM
	T12 V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13 H ₃ BO ₃ MnSO ₄ , 4H ₂ O	31 g for 1L 22,3 g for 1L	0.05 mM 0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with mili-Q water
- Adjust **T12** to **T13** to 1000 mL with mili-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8 Salt medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	10
B	10
C	10
Salt Solution	377
Trace element solution	1

- Adjust to 1000 mL with mili-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Note : This recipes corresponds to a salinity of 33g/L NaCl

Z8 Salt + saline solution a/3 medium (Z8 Salt + a/3)

(Rippka 1988)

Hypersaline cyanobacteria

Preparation of stock solutions: **Solution A, B, C, Trace element, Salt solution and Solution saline a**

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration
A	NaNO ₃	46.7	5.49 mM
	Ca(NO ₃) ₂ , 4H ₂ O or Ca(NO ₃) ₂	5.9 4.1	0.25 mM 0.25 mM
	MgSO ₄ , 7H ₂ O	2.5	0.10 mM
B	K ₂ HPO ₄ or K ₂ HPO ₄ , 3H ₂ O	3.1 4.1	0.18 mM 0.18 mM
	Na ₂ CO ₃	2.1	0.20 mM
	Salt Solution	NaCl	87.5
MgSO ₄ , 7H ₂ O		37.5	29 mM
Saline solution a	NaCl	250	0,71 M
	MgCl ₂ , 6H ₂ O	20	0,02 M
	KCl	5	0,01 M
	NaNO ₃	3	0,01 M
	MgSO ₄ , 7 H ₂ O	35	0,02 M
	CaCl ₂	5	0,01 M

For stock solutions **A, B, Salt solution** and **Solution saline a**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component		Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1	Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2	(NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3	KBr	1,21	1.02 µM
	T4	KI	0,83	0.50 µM
	T5	ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6	Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7	Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8	CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9	(NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10	Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11	Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O (ou KAl(SO ₄) ₂ , 12H ₂ O)	4,74	0.50 µM
	T12	V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13	H ₃ BO ₃ MnSO ₄ , 4H ₂ O	31 g for 1L 22,3 g for 1L	0.05 mM 0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with mili-Q water
- Adjust **T12** to **T13** to 1000 mL with mili-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8 Salt + a/3 medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	5
B	5
C	5
Trace element solution	0.5
Solution saline a	166
Salt Solution	188.5

- Adjust to 1000 mL with mili-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Z8 – Instant Ocean® medium (Z8 - 10)

Seawater cyanobacteria

Preparation of stock solutions: **Solution A, B, C and Trace element**

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration mM
A	NaNO ₃	46.7	5.49
	Ca(NO ₃) ₂ , 4H ₂ O or Ca(NO ₃) ₂	5.9 4.1	0.25 0.25
	MgSO ₄ , 7H ₂ O	2.5	0.10
B	K ₂ HPO ₄ or K ₂ HPO ₄ , 3H ₂ O	3.1 4.1	0.18 0.18
	Na ₂ CO ₃	2.1	0.20

For stock solutions **A** and **B**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component	Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1 Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2 (NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3 KBr	1,21	1.02 µM
	T4 KI	0,83	0.50 µM
	T5 ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6 Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7 Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8 CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9 (NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10 Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11 Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O	4,74	0.50 µM
	T12 V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13 H ₃ BO ₃ MnSO ₄ , 4H ₂ O	31 g for 1L 22,3 g for 1L	0.05 mM 0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with mili-Q water
- Adjust **T12** to **T13** to 1000 mL with mili-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8-IO medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	10
B	10
C	10
Trace element solution	1
Instant Ocean Salt®	20 to 33 g
NaHCO₃	100 mg (1,19mM)

- Adjust to 1000 mL with mili-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Notes:

- You can adjust the salt concentration between 20 to 33 g/L depending on the required salinity.

Z8 Mayotte medium (Z8 M)

Seawater cyanobacteria

Preparation of stock solutions: [Solution A, B, C and Trace element](#)

N° solution Stock	Component	Mass (g) for 1 L	Final molar concentration mM
A	NaNO ₃	46.7	5.49
	Ca(NO ₃) ₂ , 4H ₂ O	5.9	0.25
	or Ca(NO ₃) ₂	4.1	0.25
	MgSO ₄ , 7H ₂ O	2.5	0.10
B	K ₂ HPO ₄	3.1	0.18
	or K ₂ HPO ₄ , 3H ₂ O	4.1	0.18
	Na ₂ CO ₃	2.1	0.20

For stock solutions **A** and **B**

- Adjust to 1000 mL with mili-Q water
- Autoclave at 121°C for 15-20 minutes

N° solution Stock	Component	Mass (g) for 100 mL	Final molar concentration mM
C	C1 FeCl ₃ , 6H ₂ O	2.80	0.01
	C2 EDTA : Ethylene diamine tetra-acetate xNa ₂ x 2H ₂ O	3.9	0.01

For stock solutions **C**

- Adjust **C1** and **C2** to 100 mL with mili-Q water, for **C2** microwave heating to dissolve
- Add 10 mL of **C1** and 9.5 mL of **C2** adjust to 1000 mL with mili-Q water
- Do not autoclave the **C1** and **C2** solution, keep at 4°C

Stock solution	Component	Mass (g) for 100 mL	Final molar concentration
Trace element Solution	T1 Na ₂ WO ₄ , 2H ₂ O	0,33	0.10 µM
	T2 (NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	0,88	0.07 µM
	T3 KBr	1,21	1.02 µM
	T4 KI	0,83	0.50 µM
	T5 ZnSO ₄ , 7H ₂ O	2,87	1.00 µM
	T6 Cd (NO ₃) ₂ , 4H ₂ O	1,55	0.50 µM
	T7 Co (NO ₃) ₂ , 6H ₂ O	1,46	0.50 µM
	T8 CuSO ₄ , 5H ₂ O	1,25	0.50 µM
	T9 (NH ₄) ₂ Ni(SO ₄) ₂ , 6H ₂ O	1,98	0.50 µM
	T10 Cr(NO ₃) ₃ , 9H ₂ O	0,41	0.10 µM
	T11 Al ₂ (SO ₄) ₃ K ₂ SO ₄ , 24H ₂ O	4,74	0.50 µM
	T12 V ₂ O ₅	0,089 g for 1L	0.05 µM
	T13 H ₃ BO ₃ MnSO ₄ , 4H ₂ O	31 g for 1L 22,3 g for 1L	0.05 mM 0.01 mM

For stock solutions **Trace element Solution**

- Adjust **T1** to **T11** to 100 mL with milli-Q water
- Adjust **T12** to **T13** to 1000 mL with milli-Q water
- Add 1 mL of **T1** to **T11** and 10 mL of **T12** to **T13** adjust to 1000 mL with milli-Q water
- Autoclave at 121°C for 15-20 minutes

Preparation of Z8 M medium with stock solutions:

N° solution Stock	Volume (mL) for 1 L
A	10
B	10
C	10
Trace element solution	1
NaCl	20 g (342mM)

- Adjust to 1000 mL with milli-Q water
- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M) ou HCl (1M)
- Autoclave at 121°C for 15-20 minutes

Notes:

- You can adjust the salt concentration between 20 to 40 g/L depending on the required salinity.

Blue-Green medium (BG11/BG11₀)

(Sendersky & al, 2017, modified by C. Yepremian, this study)

Preparation of stock solutions : **Solution N°1, 2, 3, Metal trace solution (4), Vitamin solution (5)**

N° solution Stock	Component	Mass (g) for 200 mL	Molar concentration	Notes
1	MgSO ₄ . 7H ₂ O	1.3	0.284 mM	Don't autoclave !
	CaCl ₂ . 2H ₂ O	0.72	0.245 mM	
2	K ₂ HPO ₄ . 3H ₂ O	0.80	0.175 mM	Storage 3 months at 4°C * *Duration of use according to the Institut Pasteur PCC Collection (2021)
	+ EDTA (K ₂ , Mg)	0.02	2.26 μM	
	or EDTA (Na ₂)	0.02	2.26 μM	
3	Citric Acid	0.12	31.2 μM	Storage 10 days in the dark *
	+ Ammonium Ferric Citrate Green	0.12	30.0 μM	

Stock solution	Component	Mass (mg) for 200 mL	Molar concentration	Notes
Metal trace solution (4)	H ₃ BO ₃	572	46.3 μM	Storage 2 years at 4°C*
	MnCl ₂ . 4H ₂ O	362	9.15 μM	
	Zn SO ₄ . 7H ₂ O	44	0.765 μM	
	Na ₂ MOO ₄ . 2H ₂ O	78	1.61 μM	
	CuSO ₄ . 5H ₂ O	4	0.8 μM	
	CO(NO ₃) ₂ . 6H ₂ O	10	0.17 μM	

Stock solution	Component	Mass (mg) for 200 mL	Molar concentration	Notes
Vitamin solution (5) (optional)	Cyanocobalamin (B12)	8		Vitamin solution n°7 of JM/JM2 media , vitamins are dissolved in dH ₂ O/ultrapureWater pH = 4
	Thiamine HCl (B1)	8		
	Biotin	8		

For the 5 stock solutions:

- Adjust to **200** mL with dH₂O/ultrapureWater

For the stock solution **2, 3** and **Metal trace solution (4)** and **Vitamin solution (5)**:

- Autoclave at 121°C for 20 minutes

Preparation of BG11 & BG11₀ medium with stock solutions:

- To **950 mL** dH₂O/ultrapureWater ,
- **For BG11** : add 1.5g of NaNO₃(17.6 mM) ,
- **For BG11₀**: 1.03g of NaCl (17.6mM)

N° solution Stock	Volume (mL) for 1 L
1	10
2	10
3	10
Metal trace solution (4)	1
Vitamin solution (5)	1
HEPES (5mM)	1.19g

- Adjust pH=7.30 with \approx 2mL NaOH (1M)
- Adjust to 1L with dH₂O/ultrapureWater
- Autoclave at 121°C for 15- 20 minutes

For BG11 solid culture media, standard procedures can be used

For delicate strains presenting poor growth with solid media apply the following steps

- Prepare a 2x concentrated BG11 media
- Prepare a 2x concentrated agar solution (6-15g/L final) Agar can be easily purified by 2-3 decantations in dH₂O/ultrapureWater
- Autoclave at 121°C for 15- 20 minutes
- Mix BG11 media and agar solution, around 55-60°C,
- Add 10 mL 0,5 M of NaHCO₃ filtered solution (0,20 μ m), 420 mg/L solid BG11 (5mM final), mix,
- Add Na₂S₂O₃, 5H₂O (1mM final) filtered solution: 0.249 g/L (1mL 1M)
- Don't wait too long before pouring the plates!

Preparation of BG11₀:

- These are the same stock solutions as for the BG11 medium, but no NaNO₃ is added.
- A filtered solution of NaHCO₃, 420 mg/L (5 mM) can be added to partially compensate for the non-supply of sodium by NaNO₃, or 1.03 g of NaCl for a total compensation.
- For agar media, add **after** autoclaving, a **filtered** solution of NaHCO₃, 420 or even 840 mg/L (5-10 mM final) at 55-60 °C

Notes:

- According to the recipe Sendersky & al, 2017, with two main changes for the buffer concentration and the pH value.
- The main difference of BG11 from other culture media is its high nitrogen concentration and the apparent contradiction of its use by a group capable of fixing atmospheric nitrogen. This medium is compatible for the culture at high density of green microalgae such as the genera *Scenedesmus* or *Chlorella*. For some strains, the nitrogen concentration can be significantly reduced; BG11₀ media will then serve as a reference +X mM of NaNO₃. BG11 has an "ordinary" phosphorus concentration, so it is weakly buffered, cf. Rippka (1988), hence the possible addition of a buffer molecule (HEPES at \approx 5 mM). Regarding the trace solution, Rippka (1988) show that the high copper concentration which could be inhibiting for delicate species, hence a possible reduction. To date, the current PCC recipe still presents that "excess".

Bold's Basal medium (BB_{1N}/BB_{3N})

Andersen (2005), modified by Claude Yepremian, this study)

Freshwater algae

The original recipe is extremely rich in Phosphorous and in certain trace elements as Zn and Cu, our recipe practically does not modify the concentration of the macro elements, but decrease the toxicity provided by the metallic excesses

Preparation of stock solutions: [Solution A, B and Trace element solution](#)

solution Stock	Component	Mass (g) for 200 mL	Final molar concentration
A	NaNO ₃	5.0/15 BB _{1N} /BB _{3N}	2.94/8.82 mM
	CaCl ₂ · 2H ₂ O	0.5	0.17 mM
	MgSO ₄ · 7H ₂ O	1.5	0.30 mM
	Fe-EDTA	0.2	27.24 μM
B	K ₂ HPO ₄ or K ₂ HPO ₄ · 3H ₂ O	1.5 or 1.96	0.43 mM 0.43 mM
	KH ₂ PO ₄	3.5	1.29 mM
	NaCl	0.4	0.34 mM

Stock solution	Component	Mass (mg) for 200 mL	Final molar concentration
Trace element solution	H ₃ BO ₃	480	39.00 μM
	MnCl ₂ · 4H ₂ O	360	9.00 μM
	(NH ₄) ₆ Mo ₇ O ₂₄ · 4H ₂ O	20	0.08 μM
	ZnSO ₄ · 7H ₂ O	44	0.76 μM
	CuSO ₄ · 5H ₂ O	3.2	0.06 μM
	CoSO ₄ · 7H ₂ O or CoSO ₄ · H ₂ O	18 11	0.32 μM 0.32 μM
	VO ₂ SO ₄ · 2H ₂ O	8.6	0.22 μM

For stock solutions **A**, **B** and **Trace element solution**:

- Adjust to **200 mL** with dH₂O/ultrapure Water
- Autoclave at 121°C for 15-20 minutes

Preparation of BB_{1N/b3} medium with stock solutions:

- To **950 mL** dH₂O/ultrapure Water, add the amounts of the 3 stock solutions , complete to 1L

Solution Stock	Volume (mL) for 1 L
A	10
B	10
Trace element solution	1

- Optional: pH can be adjusted to 7 with about 0,5mL NaOH (1M), without adjustment pH≈6,4-6,55
- Autoclave at 121°C for 15-20 minutes

Notes :

- For solid culture media, add 12-15 g/L of agar
- The addition of vitamin solution can be beneficial. The solution number 7 of JM media can be used, (<http://www.ccap.ac.uk/media/documents/JM.pdf>), it can be added before or after autoclaving (previously sterilized by filtration on 0.22µm)
- A triple nitrogen concentration, compared with B_{1N} medium, may be advantageous to obtain dense cultures of *Chlorella* or *Scenedesmus*: BB_{3N}

DIATOM Medium (DM2)

(CCAP2021, modified by Claude Yepremian. Yepremian, this study)

Freshwater Diatoms

Preparation of stock solutions : **Solution N°1, 2, 3, 4, 5, 6, 7, 8**

N° stock solution	Component	Mass (g) for 200 mL	Molar concentration in final solution
1 (JM solution)	Ca(NO ₃) ₂ . 4H ₂ O	4	0.17 mM
2(JM solution)	KH ₂ PO ₄	2.48	91.12 µM
3(JM solution)	MgSO ₄ . 7H ₂ O	10	0.10 mM
4(JM solution)	NaHCO ₃	3.18	0.19 mM
5(JM solution)	EDTA FeNa	0.45	6.13 µM
	EDTA Na ₂	0.45	6.04 µM
6 (JM solution)	H ₃ BO ₃	0.496	40.13 µM
	MnCl ₂ . 4H ₂ O	0.278	7.02 µM
	(NH ₄) ₆ MO ₇ O ₂₄ . 4H ₂ O	0.200	0.81 µM
7 (JM solution)	Cyanocobalamin	0.008	0.03 µM
	Thiamin HCl	0.008	0.12 µM
	Biotin	0.008	0.16 µM
8 (L1 medium)	Na ₂ SiO ₃ . 9H ₂ O	6	0.21 mM

For the **8 stock solutions**:

- Adjust to **200 mL** with dH₂O/ultrapure Water,
- Autoclave stock solutions at 121°C for 15-20 minutes

Preparation of JM2 medium from stock solutions:

- To **950 mL** dH₂O/ultrapure Water, add the amounts of the 8 stock solutions ,
- Add 0.42 g MOPS (acid form, 2 mM), adjust pH to 6.9 with NaOH 5M, complete to 1L

N° stock solution	Volume (mL) for 1L
1	2
2	1
3	0.5
4	1
5	1
6	1
7	1
8	2

- Autoclave at 121°C for 15-20 minutes,
- For solid medium: Agar concentration 12g/L

Note :

Medium very poor for nitrogen and phosphorous, very poorly buffered also, explaining the addition of MOPS, by comparison with the original recipe, we also doubled the nitrogen concentration

Dunaliella medium

Halophilic algae

Borowitzka (1988), modified by Claude Yepremian, MNHN)

Preparation of stock solutions: [Solution A, B, C, D and Trace element](#)

solution Stock	Component	Mass (g) for 200 mL	Final molar concentration
A	KNO ₃	20	9.89mM
	CaCl ₂ /CaCl ₂ , 2H ₂ O	4/5.3	1.80 mM
	KCl	4	2.68 mM
	Fe EDTA	0,4	54.50 μM
B (Medium BB_{1M/3M})	K ₂ HPO ₄	1.5	0.43 mM
	or K ₂ HPO ₄ , 3H ₂ O	or 1.96	
	KH ₂ PO ₄	3.5	1.29 mM
	NaCl	0.4	0.34 mM
C	MgCl ₂ , 6 H ₂ O	30 g	7.38 mM
	MgSO ₄ , 7 H ₂ O	10 g	2.03 mM
D	NaHCO ₃	8.6 g	0.51 mM

Stock solution	Component	Mass (mg) for 200 mL	Final molar concentration
Trace element Solution 1 (Medium BB_{1N}/BB_{3N})	H ₃ BO ₃	480	39.00μM
	MnCl ₂ , 4H ₂ O	360	9.00μM
	(NH ₄) ₆ Mo ₇ O ₂₄ , 4H ₂ O	20	0.08 μM
	ZnSO ₄ , 7H ₂ O	44	0.76 μM
	CuSO ₄ , 5H ₂ O	3.2	0.06 μM
	CoSO ₄ , 7H ₂ O	18	0.32 μM
	or CoSO ₄ , 1 H ₂ O	11	
VOSO ₄ , 2H ₂ O	8.6	0.22 μM	

L1 Trace element solution

- **To 950 mL** dH₂O/ultrapureWater, after dissolving EDTA and Iron add the amounts of the 9 primary stock solutions , complete to 1L, Autoclave at 121°C for 15-20 minutes

	Component	Primary Stock Solutions	Quantity	Final molar concentration
Trace element Solution 2 (Medium L1)	Na ₂ EDTA ·	---	4.36 g	2.34 µM
	FeCl ₃ ·	---	3.15 g	2.34 µM
	MnCl ₂ ·4	1.780 g /100mL dH ₂ O	10 mL	180 nM
	ZnSO ₄ ·	0.230 g/100mL dH ₂ O	10 mL	16 nM
	CoCl ₂ ·	1.190 g/100mL dH ₂ O	1 mL	10 nM.
	CuSO ₄ ·	0.250 g/100mL dH ₂ O	1 mL	2 nM
	Na ₂ MoO ₄ · 2H ₂ O	0.199 g/100mL dH ₂ O	10 mL	16 nM
	H ₂ SeO ₃	0.129 g/100mL dH ₂ O	1 mL	2 nM
	NiSO ₄ ·	0.263 g/100mL dH ₂ O	1 mL	2 nM
	Na ₃ VO ₄	0.184 g/100mL dH ₂ O	1 mL	2 nM
	K ₂ CrO ₄	0.194 g/100mL dH ₂ O	1 mL	2 nM

solution Stock	Component	Mass (g) for 200 mL	Final molar concentration
Vitamin solution (Solution 7 medium JM2)	Cyanocobalamin	0.008	0.03 µM
	Thiamin HCl	0.008	0.12 µM
	Biotin	0.008	0.16 µM

Preparation of *Dunaliella* medium with stock solutions:

- **To 800 mL** dH₂O/ultrapure Water, add the amounts of NaCl requested and the amounts of the 7 stock solutions, add TRIS base, adjust pH to **7.50** with HCl 6 M, complete to 1L, Autoclave at 121°C for 15-20 minutes

Solution Stock	Volume (mL) for 1 L
A	10 mL
B	2 mL
C	10 mL
D	1mL/2.7ml*
Trace element solution 1 (BB)	1 mL
Trace element solution 2 (L1)	0.2 mL
Vitamin solution (JM2)	1 mL
TRIS base (2.5 mM)	0.305 g

*if solution 4 of culture medium JM2 is used

- For solid culture media, add 12-15 g/L of agar

HEMERICK medium

Porphyridium brackish microalgae

Hemerick (1973), modified by Claude Yepremian, MNHN)

Preparation of stock solutions: **Solution A, B and Trace element**

Stock solution	Component	Mass (g) for 200 mL	Final molar concentration
A	NaNO ₃	34 g	4 mM
	CaCl ₂ ·2H ₂ O	29,4	2mM
	KCl	15 g	2 mM
B	K ₂ HPO ₄	3.5	

Stock solution	Component	Mass (mg) for 200 mL	Final molar concentration
Trace element Solution(BB1N/BB3N)	H ₃ BO ₃	480	39.00 μM
	MnCl ₂ ·4H ₂ O	360	9.00 μM
	(NH ₄) ₆ Mo ₇ O ₂₄ ·4H ₂ O	20	0.08 μM
	ZnSO ₄ ·7H ₂ O	44	0.76 μM
	CuSO ₄ ·5H ₂ O	3.2	0.06 μM
	CoSO ₄ ·7H ₂ O or CoSO ₄ ·1 H ₂ O	18 11	0.32 μM
	VOSO ₄ ·2H ₂ O	8.6	0.22 μM

For stock solutions **A, B** and **Trace element solution**:

- Adjust to 200 mL with dH₂O/ultrapure Water
- Autoclave at 121°C for 15-20 minutes

Preparation of BB_{1N/b3} medium with stock solutions:

- **To 950 mL** dH₂O/ultrapure Water, add after weighting :
- 29 g NaCl (0.5 M)
- 12.3 g MgSO₄·7H₂O (50 mM)
- 50 mg FeEDTA (0.14 mM)
- add the amounts of the 3 stock solutions , adjust pH to 7, complete to 1L

Stock Solution	Volume (mL) for 1 L
A	10
B	10
Trace element solution	1

Notes:

- The addition of vitamin solution can be beneficial. The solution number 7 of JM media can be used, (<http://www.ccap.ac.uk/media/documents/JM.pdf>), it can be added before or after autoclaving (previously sterilized by filtration on 0.22µm)

Jaworski's Medium (JM and JM2)

CCAP(2021), slightly modified by Claude Yepremian, this study)

Freshwater algae

Preparation of stock solutions: [Solution N°1, 2, 3, 4, 5, 6, 7, 8, 9](#)

N° stock solution	Component	Mass (g) for 200 mL	Molar concentration in final medium
1	Ca (NO ₃) ₂ . 4H ₂ O	4.000	84.69 µM
2	KH ₂ PO ₄	2.480	91.12 µM
3	MgSO ₄ . 7H ₂ O	10.000	0.20 mM
4	NaHCO ₃	3.180	0.19 mM
5	EDTA FeNa	0.450	6.13 µM
	EDTA Na ₂	0.450	6.04 µM
6	H ₃ BO ₃	0.496	40.13 µM
	MnCl ₂ . 4H ₂ O	0.278	7.02 µM
	(NH ₄) ₆ MO ₇ O ₂₄ . 4H ₂ O	0.200	0.81 µM
7	Cyanocobalamin	0.008	0.03 µM
	Thiamin HCl	0.008	0.12 µM
	Biotin	0.008	0.16 µM
8	NaNO ₃	16.000	0.94 mM
9	Na ₂ HPO ₄ . 12H ₂ O	7.200	0.10 mM

For the 9 stock solutions:

- Adjust to **200 mL** with dH₂O/ultrapureWater
- Autoclave stock solutions at 121°C for 15-20 minutes

Preparation of JM2 medium from stock solutions:

- To **950 mL** dH₂O/ultrapure Water, add the amounts of the 9 stock solutions , complete to **1L**

N° stock solution	Volume (mL) for 1L	Molar concentration in
1	1	
2	1	
3	1	
4	1	
5	1	
6	1	
7	1	
8	2	
9	1	

- Autoclave at 121°C for 15-20 minutes, nominal pH ≈ 7.40
- For solid medium: Agar concentration 12g/L.

Notes :

- One of the most used freshwater media, tolerated by distinct algal groups, probably by the fact of its oligotrophic character, recommended to be used for isolation step
- The only difference between **JM2** medium and **JM** medium lies in the doubling of the concentration of Na NO₃, 2mL instead of 1mL per liter.

L1 medium

CCAP (2021), NCMA (2021), modified by Claude Yepremian, this study)

Marine algae

Preparation of stock solutions : **L1 Trace element solution and Vitamin solution**

Stock solution	Component	Primary Stock Solutions mass (g) for 100mL	Quantity for 1L	Final molar concentration
L1 Trace element solution	Na ₂ EDTA. 2H ₂ O	---	4.36 g	11.70 μM
	FeCl ₃ . 6H ₂ O	---	3.15 g	11.70 μM
	MnCl ₂ . 4H ₂ O	1.780	10 mL	0.90 μM
	ZnSO ₄ . 7H ₂ O	0.230	10 mL	0.08 μM
	CoCl ₂ . 6H ₂ O	1.190	1 mL	0.05 μM
	CuSO ₄ . 5H ₂ O	0.250	1 mL	0.01 μM
	Na ₂ MoO ₄ . 2H ₂ O	0.199	10 mL	0.08 μM
	H ₂ SeO ₃	0.129	1 mL	0.01 μM
	NiSO ₄ . 6H ₂ O	0.263	1 mL	0.01 μM
	Na ₃ VO ₄	0.184	1 mL	0.01 μM
	K ₂ CrO ₄	0.194	1 mL	0.01 μM

For the **L1 Trace element solution** :

- To **950 mL** dH₂O/ultrapureWater, after dissolving EDTA and Iron add the amounts of the 9 primary stock solutions , complete to 1 L.
- Autoclave at 121°C for 15-20 minutes

Stock solution	Component	Primary Stock Solutions mass (g) for 100mL	Quantity for 1L	Final molar concentration
Vitamin solution	Thiamine · HCl (B1)	---	200 mg	0.296 μM
	Biotin (H)	0.1	1 mL	2.050 nM
	Cyanocobalamin (B12)	0.1	1 mL	0.369 nM

For the **Vitamin Solution** :

- **To 1 L** dH₂O/ultrapureWater first dissolve the 200 mg Thiamine, add the aliquots of Biotin and Cyanocobalamin solutions
- Autoclave at 121°C for 15-20 minutes

Preparation of L1 medium with stock solutions:

Solution Stock	Component	Mass (g) for 100mL	Quantity	Final molar concentration
1	NaNO ₃	7.5	1 mL	0.88 mM
2	NaH ₂ PO ₄ , H ₂ O	0.5	1 mL	36.20 μM
3	Na ₂ SiO ₃ , 9H ₂ O	3	1 mL	0.105 mM
L1 Trace element solution	---	---	1 mL	---
Vitamin solution	---	---	0.5mL	---

- Add the **five stocks solutions** to **1 L** artificial seawater (prepared by dissolving: 32 g Crystal Reef® or Instant Ocean® in 1 L dH₂O/ultrapureWater)
- Adjust the pH to 7.80-8.00, Autoclave at 121°C for 15-20 minutes

Notes :

- The NCMA recipe uses natural sea water. As the majority of the marine algae recipes, the medium L1 is poor in nitrogen and phosphorous, it presents a high diversity in trace metals; that is probably what makes the difference from the popular f/2, in terms of performance of culture.

SPIRULINA medium

Aiba & Ogawa (1977), modified by Claude Yepremian, this study)

Alcaline waters microalgae

Preparation of stock solutions: [Solution A, B, C, Trace element \(1\) Trace element \(2\), Vitamin solution](#)

Stock solution	Component	Mass (g) for 200 mL	Final molar concentration
A	NaNO ₃	50	29.41 mM
	MgSO ₄ , 7H ₂ O	4	0.81 mM
	CaCl ₂ , 2H ₂ O	0.8	0.27 mM
B	K ₂ SO ₄	20	5.74 mM
	NaCl	20	17.11mM
		Stir and heat to dissolve(≈ 70°C)	

Stock solution	Component	Mass (g) for 200 mL	Final molar concentration
C	Na ₂ EDTA · 2H ₂ O	1.68 g Heat to dissolve by microwave (≈ 70°C)	250 μM
	FeSO ₄ , 7H ₂ O	0.27 g	48.6 μM

Stock solution	Component	Mass (g) for 1000 mL	Final molar concentration
D	NaHCO ₃	68.05	0.162 M
	Na ₂ CO ₃	20.15	38.02 mM
	K ₂ HPO ₄	2.50	2.88 mM

Stock solution	Component	Mass (mg) for 200 mL	Final molar concentration
Trace element n° 1 Medium(BB_{1N}/BB_{3N})	H ₃ BO ₃	480	39.00μM
	MnCl ₂ , 4H ₂ O	360	9.00μM
	(NH ₄) ₆ Mo ₇ O ₂₄ ,4H ₂ O	20	0.08 μM
	ZnSO ₄ ,7H ₂ O	44	0.76 μM
	CuSO ₄ ,5H ₂ O	3.2	0.06 μM
	CoSO ₄ ,7H ₂ O or CoSO ₄ ,1 H ₂ O	18 11	0.32 μM
	VO ₂ SO ₄ ,2H ₂ O	8.6	0.22 μM

L1 Trace element solution n°2

- To 950 mL dH₂O/ultrapureWater, after dissolving EDTA and Iron add the amounts of the 9 primary stock solutions , complete to 1L, Autoclave at 121°C for 15-20 minutes

Component	Primary Stock Solutions	Quantity	Final molar concentration
Na ₂ EDTA · 2H ₂ O	---	4.36 g	2.34 μM
FeCl ₃ · 6H ₂ O	---	3.15 g	2.34 μM
MnCl ₂ ·4 H ₂ O	1.780 g /100mL dH ₂ O	10 mL	180 nM
ZnSO ₄ · 7H ₂ O	0.230 g/100mL dH ₂ O	10 mL	16 nM
CoCl ₂ · 6H ₂ O	1.190 g/100mL dH ₂ O	1 mL	10 nM.
CuSO ₄ · 5H ₂ O	0.250 g/100mL dH ₂ O	1 mL	2 nM
Na ₂ MoO ₄ · 2H ₂ O	0.199 g/100mL dH ₂ O	10 mL	16 nM
H ₂ SeO ₃	0.129 g/100mL dH ₂ O	1 mL	2 nM
NiSO ₄ · 6H ₂ O	0.263 g/100mL dH ₂ O	1 mL	2 nM
Na ₃ VO ₄	0.184 g/100mL dH ₂ O	1 mL	2 nM
K ₂ CrO ₄	0.194 g/100mL dH ₂ O	1 mL	2 nM

Stock solution	Component	Mass (g) for 200 mL	Final molar concentration
Vitamin Solution n° 7 (medium JM/JM2)	Cyanocobalamin	0.008	0.03 μM
	Thiamin HCl	0.008	0.12 μM
	Biotin	0.008	0.16 μM

Preparation of Spirulina medium with stock solutions:

SOLUTION 1	In a 1 Liter Duran bottle, To 300 ml of dH ₂ O/ultrapure water add : 200 mL of solution D
SOLUTION 2	In a 0.5 Litre Duran bottle, to 468-470 ml of dH ₂ O/ultrapure water add successively: <ul style="list-style-type: none"> • 10 mL solution A • 10 mL solution B • 10 mL solution C • 1 mL Trace Solution n°1 (medium BB) • Optionnal, 0.2 mL Trace Solution n°2 medium L1) • 1 mL Vitamins solution (medium JM/JM2)

Autoclave solutions 1&2 separately, 15-20 min at 121° C

Aseptically combine the 2 solutions, under PSM per ex.

Notes:

- The original recipe is extremely rich in Phosphorous, nitrogen as well as in HCO₃⁻/CO₃²⁻, our recipe practically does not modify the concentration of these macro elements

TAP(mixotrophic/heterotrophic)TP(autotrophic) media

CHLAMYCOLLECTION (2021), modified b Claude Yepremian, this study)

Chlamydomonas species Freshwater algae

TAP is the most popular medium to cultivate the model organism *Chlamydomonas reinhardtii* under a mixotrophic metabolism. The original recipe is time consuming to prepare.

Preparation of stock solutions: **Solution A, B and C**

Solution Stock	Component	Mass (g) for 100 mL	Final molar concentration
A	Tris base	24.20 g	20 mM
	Glacial acetic acid	13.64g/13 mL	17.5 mM
B	NH ₄ Cl	4.00 g	7.5 mM
	MgSO ₄ . 7H ₂ O	1.00 g	0.41 mM
	CaCl ₂ . 2H ₂ O	0.50 g	0.34 mM
C (nominal pH : 7.2)	K ₂ HPO ₄	10,80 g	0.62 mM
	KH ₂ PO ₄	5,60 g	0.41 mM

Note : **Solution B** : NH₄Cl 400 mg/L; **Solution C** : K₂HPO₄+ KH₂PO₄ 164 mg/L final

Preparation of stock solution: **Trace element solution (Filner's recipe)**

Dissolve 5g Na₂EDTA, 2H₂O by heating (60-70°C) in 400 mL dH₂O/ultrapure Water, adjust the pH with KOH 2N to 6.5-6.8 (≈ 5 mL) , add successively the other trace elements after complete dissolution with each.

Stock solution	Component	Mass (mg) for 500 mL	Final molar concentration
Trace element solution Modified Hutner's (Filner's recipe)	Na ₂ EDTA. 2H ₂ O	5000	134.0 μM
	FeSO ₄ . 7H ₂ O	500	18.0 μM
	ZnSO ₄ . 7H ₂ O	2200	136.0 μM
	H ₃ BO ₃	1140	184.0 μM
	MnCl ₂ . 4H ₂ O	510	25.8 μM
	CuSO ₄ .5H ₂ O	160	6.4 μM
	Na ₂ MoO ₄ . 2H ₂ O or H ₂ MoO ₄	98 73	4.1 μM
	CoCl ₂ . 6H ₂ O	160	6.7 μM

For stock solutions **A, B** and **Trace element solution**:

- Adjust to 100 or 500 mL with dH₂O/ultrapure Water
- Autoclave at 121°C for 15-20 minutes

Preparation of TAP/TP medium with stock solutions:

- To **950 mL** dH₂O/ultrapure Water, add the amounts of the 4 stock solutions , complete to 1L

Solution Stock	Volume (mL) for 1 L
A	10
B	10
C	1
Trace element solution	5

- Adjust pH to 7 with about 0.7 mL NaOH (5M),
- Autoclave at 121°C for 15-20 minutes

To prepare the autotrophic medium: TP, prepare a stock solution A where Glacial acetic acid is omitted , adjust the pH of the final solution with HCL 6 N

- For solid culture media, add 12-15 g/L of agar

Notes :

- The unique nitrogen source is Ammonium, some trace elements notably Zn are present at high concentrations. The initial dilution of the modified Hutner's trace element allows to avoid the so laborious preparation of the original recipe. On the other hand, that allows an increase in the concentration of some metallic elements in the medium which could be noxious.