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(Modified from: Chen Y. B., Zehr J. P., Mellon M., 1996. Growth and nitrogen fixation of the diazotrophic filamentous non-heterocystous cyanobacterium *Trichodesmium* sp. IMS 101 in defined media: evidence for a circadian rhythm . *J Phycol.* 32: 916-923.)

Minerals	Stock Solutions (g/L)	Quantity (mL Stock/L Media)	Final Concentration (mM)
NaCl	245.45	100	420
MgCl ₂ · 6 H ₂ O	406.6	10	20
KCl	74.60	10	10
MgSO ₄ · 7 H ₂ O	603.8	10	25
CaCl ₂ · 2 H ₂ O	147	10	10

Adjust to 900mL with mQ water and autoclave.

After cooling, add the following filter sterilized (0.2 µm) components to complete the medium:

Minerals	Stock Solutions (g/L)	Quantity (mL Stock/L Media)	Final Concentration (mM)
NaNO ₃	150	10	16
NaHCO ₃	21	10	2.5
K ₂ HPO ₄ · 3H ₂ O	6.8	1	0.03
Na ₂ CO ₃	26.5	0.6	0.15
Fe-NH ₄ -citrate	6	0.25	-
KBr	115.7	1	0.97
NaF	2.9	1	0.07
Trace Metal Mix 4	See recipe below	1	-
Trace Metal Mix	See recipe below	1	-
Vitamins3 Mix	See recipe below	1	-

Check the pH, has to be between 8.12 and 8.2.

For solid medium use 7g/L of agarose. Sterilize the agarose separately in 550 ml of milliQ water. In this case the mineral solution is filled up to 400 ml.

Trace Metal Mix 4:

Trace metals	Quantity g/L	Concentration in the final media (mM)
H ₃ BO ₃	35.9	0.0006
SrCl ₂ · 6H ₂ O	17.3	0.00006
LiCl	1.1	0.00003
Na ₂ SeO ₃ · 5H ₂ O	0.5mL of a stock of 32mg/L	0.00000006

Trace Metal Mix:

Trace metals	Stock 1 (g/100mL)	Trace metal mix (Stock1 mL/L)	Concentration in the final media (mM)
EDTA	-	0.74 g	2.5
FeCl ₃ · 6H ₂ O	-	0.11 g	0.0004
MnCl ₂ · 4H ₂ O	0.4	1	0.00002
ZnSO ₄ · 7H ₂ O	0.12	1	0.000004
CoCl ₂ · 6H ₂ O	0.06	1	0.000002
Na ₂ MoO ₄ · 2H ₂ O	0.27	1	0.00001
CuSO ₄ · 5H ₂ O	0.025	1	0.000001

Vitamins 3 Mix:

Vitamins	Vitamins 3 mix (quantity/100mL)
Thiamine-HCl	10 mg
d-biotin*	100 µL (from a stock of 5 mg in 10mL)
Vitamin B12	100 µL (from a stock of 5 mg in 10mL)

*Dissolve first in 0.1 mL 2M NaOH. Then add 9.9 mL of mQ water.