## 2xASN III

Minerals	Stock Solutions (g/L)	Quantity (mL Stock/L Media)	Final Concentration (mM)
		•••	
NaCl	250	200	856
MgCl <sub>2</sub> ·6H <sub>2</sub> O	200	20	20
KCl	50	20	13
MgSO <sub>4</sub> · 7H <sub>2</sub> O	350	20	28
CaCl <sub>2</sub> ·2H <sub>2</sub> O	50	20	6
Na₃-citrate	0.6	10	0.023
Na <sub>2</sub> -EDTA <sup>-</sup> 2H <sub>2</sub> O	0.1	10	0.003
Trace metal mix (A5 + Co)	See recipe below	1	-

Adjust to 900mL with mQ water and autoclave.

After cooling, add the following components with a sterile filter (0.2  $\mu m$ ) to complete the media:

Minerals	Stock Solutions (g/L)	Quantity (mL Stock/L Media)	Final Concentration (mM)
NaNO <sub>3</sub>	150	5	8
K <sub>2</sub> HPO <sub>4</sub> ·3H <sub>2</sub> O	4	5	0.088
Na <sub>2</sub> CO <sub>3</sub>	20	1	0.19
Fe-NH <sub>4</sub> -citrate	6	0.5	-
Vitamin B12 (Cyanocobalamin)	0.02	1	-

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 A5 + Co:**

Trace metals	Quantity g/L	Concentration in the final media (mM)
H <sub>3</sub> BO <sub>3</sub>	2.86	0.047
MnCl <sub>2</sub> · 4H <sub>2</sub> O	1.81	0.009
ZnSO <sub>4</sub> · 7H <sub>2</sub> O	0.22	0.0007
Na <sub>2</sub> MoO <sub>4</sub> · 2H <sub>2</sub> O	0.39	0.0016
CuSO <sub>4</sub> · 5H <sub>2</sub> O	0.08	0.0003
$Co(NO_3)_2$ · $6H_2O$	0.05	0.0002