

## Kropat's Trace Elements Solutions

from Kropat J, Hong-Hermesdorf A, Casero D, Ent P, Castruita M, Pellegrini M, Merchant SS, Malasarn D (2011) A revised mineral nutrient supplement increases biomass and growth rate in *Chlamydomonas reinhardtii*. *Plant J.* 66:770-80

### Revised Trace Elements Recipe

Make preliminary concentrated stock solutions in Part A first, and, where indicated, use these to make the individual stock solutions in Part B listed below. Only solutions in Part B are added directly to media. **Sodium selenite ( $\text{Na}_2\text{SeO}_3$ ) is considered dangerous so handle with care. See: <https://www.sigmaaldrich.com/US/en/sds/sigma/214485>.**

#### A. Preliminary concentrated stock solutions

Pre-1. EDTA- $\text{Na}_2$ concentrate	125 mM	13.959 g in ~ 250 ml, titrate to pH 8.0 with trace element grade KOH (~1.7 g), and bring up to a volume of 300 ml
Pre-2. $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}$ concentrate	285 $\mu\text{M}$	$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}\cdot 4\text{H}_2\text{O}$ : 0.088 g, bring up to a volume of 250 mL
Pre-3. $\text{Na}_2\text{SeO}_3$ concentrate	1 mM	$\text{Na}_2\text{SeO}_3$ : 0.043 g, bring up to a volume of 250 mL

#### B. Individual Stock Solutions for medium (1000 $\times$ )

Bring each stock solution up to 250 mL in water. Use 1 mL of each individual stock solution in 1 L medium.

Stock Solution	Concentration in stock	Composition
1. EDTA- $\text{Na}_2$	25 mM	EDTA- $\text{Na}_2$ : 50 mL of 125 mM EDTA- $\text{Na}_2$ concentrate (Pre-1) from Step A
2. $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}$	28.5 $\mu\text{M}^*$	$(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}\cdot 4\text{H}_2\text{O}$ : 25 mL of 285 $\mu\text{M}$ $(\text{NH}_4)_6\text{Mo}_7\text{O}_{24}$ concentrate (Pre-2) from Step A
3. $\text{Na}_2\text{SeO}_3$	0.1 mM	$\text{Na}_2\text{SeO}_3$ : 25 mL of 1 mM $\text{Na}_2\text{SeO}_3$ concentrate (Pre-3) from Step A
4. Zn-EDTA	2.5 mM 2.75 mM	Zn $\text{SO}_4\cdot 7\text{H}_2\text{O}$ : 0.18 g EDTA- $\text{Na}_2$ : 5.5 mL of 125 mM EDTA- $\text{Na}_2$ concentrate (Pre-1) from Step A

5. Mn-EDTA	6 mM 6 mM	MnCl <sub>2</sub> ·4H <sub>2</sub> O: 0.297 g EDTA-Na <sub>2</sub> : 12 mL of 125 mM EDTA-Na <sub>2</sub> concentrate (Pre-1) from Step A
6. Fe-EDTA	20 mM 22 mM 22 mM	FeCl <sub>3</sub> ·6H <sub>2</sub> O: 1.35 g EDTA-Na <sub>2</sub> : 2.05 g Na <sub>2</sub> CO <sub>3</sub> (sodium carbonate): 0.58 g (Combine EDTA-Na <sub>2</sub> with sodium carbonate in water and mix. Add FeCl <sub>3</sub> ·6H <sub>2</sub> O after the first two components dissolve. Do Not Use Pre-1.)
7. Cu-EDTA	2 mM 2 mM	CuCl <sub>2</sub> ·2H <sub>2</sub> O: 0.085 g EDTA-Na <sub>2</sub> : 4 mL of 125 mM EDTA-Na <sub>2</sub> concentrate (Pre-1) from Step A

Notes:

\*The final [Mo] in the 1× medium is 0.2 μM

Total [EDTA] in 1× medium: (25 + 2.75 + 6 + 22 + 2) = 57.75 μM