

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.

A. Preliminary concentrated stock solutions

Pre-1. EDTA-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. (NH ₄) ₆ Mo ₇ O ₂₄ concentrate	285 μM	(NH ₄) ₆ Mo ₇ O ₂₄ ·4H ₂ O: 0.088 g, bring up to a volume of 250 mL
Pre-3. Na ₂ SeO ₃ concentrate	1 mM	Na ₂ SeO ₃ : 0.043 g, bring up to a volume of 250 mL

B. Individual Stock Solutions for medium (1000×)

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-Na ₂	25 mM	EDTA-Na ₂ : 50 mL of 125 mM EDTA-Na ₂ concentrate (Pre-1) from Step A
2. (NH ₄) ₆ Mo ₇ O ₂₄	28.5 μM*	(NH ₄) ₆ Mo ₇ O ₂₄ ·4H ₂ O: 25 mL of 285 μM (NH ₄) ₆ Mo ₇ O ₂₄ concentrate (Pre-2) from Step A
3. Na ₂ SeO ₃	0.1 mM	Na ₂ SeO ₃ : 25 mL of 1 mM Na ₂ SeO ₃ concentrate (Pre-3) from Step A
4. Zn·EDTA	2.5 mM 2.75 mM	ZnSO ₄ ·7H ₂ O: 0.18 g EDTA-Na ₂ : 5.5 mL of 125 mM EDTA-Na ₂ concentrate (Pre-1) from Step A

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