

Waris-H + Selen

| Components and Final Concentration in Culture Medium | Stock Solution | Addition per Litre of Culture Medium |
|--|---------------------------------|---|
| 1. HEPES (1.00 mM) | 238.10 g / l dH ₂ O | 1 ml |
| 2. KNO ₃ (1.00 mM) | 100.00 g / l dH ₂ O | 1 ml |
| 3. MgSO ₄ x 7 H ₂ O (81.1 μM) | 20.00 g / l dH ₂ O | 1 ml |
| 4. (NH ₄) ₂ HPO ₄ (0.15 mM) | 20.00 g / l dH ₂ O | 1 ml |
| 5. Ca(NO ₃) ₂ x 4 H ₂ O (0.42 mM) | 100.00 g / l dH ₂ O | 1 ml |
| 6. Vitamin Solution | | 3 ml |
| Vitamin B12 (0.15 nM) | 0.20 mg / l dH ₂ O | |
| Biotin (4.10 nM) | 1.00 mg / l dH ₂ O | |
| Thiamine-HCl (0.30 μM) | 100.00 mg / l dH ₂ O | |
| Niacinamide (0.80 nM) | 0.10 mg / l dH ₂ O | |
| pH of the Vitamin Solution should be around pH 7 | | |
| 7. P-II Metals | | 1 ml |
| EDTA (Titriplex III) (8.06 μM) | 3.00 g / l dH ₂ O | |
| H ₃ BO ₃ (18.43 μM) | 1.14 g / l dH ₂ O | |
| MnCl ₂ x 4 H ₂ O (0.73 μM) | 144.00 mg / l dH ₂ O | |
| ZnSO ₄ x 7 H ₂ O (73.00 nM) | 21.00 mg / l dH ₂ O | |
| CoCl ₂ x 6 H ₂ O (16.80 nM) | 4.00 mg / l dH ₂ O | |
| Dissolve EDTA and boric acid in dH ₂ O, then add metals one after the other. | | |
| 8. Fe-EDTA | | 1 ml |
| EDTA (Titriplex II) (17.86 μM) | 5.22 g / l dH ₂ O | |
| FeSO ₄ x 7 H ₂ O (17.90 μM) | 4.98 g / l dH ₂ O | |
| 1 N KOH | 54.00 ml / l dH ₂ O | |
| EDTA (Titriplex II) and FeSO ₄ x 7 H ₂ O is heated for 30 min (100°C); KOH is added to the cooled mixture. | | |
| 9. Na ₂ SeO ₃ x 5 H ₂ O | 160 mg / l dH ₂ O | 50 μl |
| 10. Soil extract | | 10 ml |

adjust the pH to 7.0 and autoclave

Preparation of Soil Extract

10 g of garden-soil is mixed with 120 ml dH₂O and boiled for 10 minutes. Afterwards it is centrifuged for 10 minutes (low speed), and the supernatant is filtered through a series of membrane filters from 1.2 μm – 0.1 μm pore size. The remaining filtrate is adjusted to 100 ml with dH₂O. Aliquots of 10 ml are stored frozen.

The soil should not be recently fertilized and should not contain too much humus.