

Sulfolobus Plates (YS or T) (*ala Zillig*) for 1 liter
(revised September 2007)

1. Dissolve in approx. 800 ml distilled water:
 - a. 3.0 grams Ammonium sulfate: $(\text{NH}_4)_2\text{SO}_4$
 - b. 0.7 grams Glycine
 - c. 0.5 grams Potassium hydrogen phosphate (potassium phosphate, dibasic): K_2HPO_4
 - d. 0.1 grams Potassium chloride: KCl

2. Carbon source:
 - YS:
 - a. 1.0 gram Yeast extract
 - b. 2.0 grams Sucrose

 - Tryptone (T):
 - a. 2.0 grams Tryptone

3. Trace elements (solutions are on shelf above the scale):
 - a. 200 μl 1% Ferrous sulfate: FeSO_4
 - b. 244 μl 1% Sodium borate: $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10 \text{H}_2\text{O}$
 - c. 90 μl Manganese chloride: $\text{MnCl}_2 \cdot 4 \text{H}_2\text{O}$
 - d. 11 μl Zinc sulfate: $\text{ZnSO}_4 \cdot 7 \text{H}_2\text{O}$
 - e. 2.5 μl Cupric sulfate: $\text{CuSO}_4 \cdot 5 \text{H}_2\text{O}$
 - f. 1.5 μl Sodium molybdate: $\text{Na}_2\text{MoO}_4 \cdot 2 \text{H}_2\text{O}$
 - g. 1.5 μl Vanadyl sulfate: $\text{VOSO}_4 \cdot 5 \text{H}_2\text{O}$
 - h. 0.5 μl Cobalt chloride: $\text{CoCl}_2 \cdot 6 \text{H}_2\text{O}$
 - i. 0.5 μl Nickel sulfate: $\text{NiSO}_4 \cdot 6 \text{H}_2\text{O}$

4. Adjust pH to 3.0 – 3.2 with 50% sulfuric acid (approx. 300 μl)

5. Add distilled water to make 1 liter

6. Dissolve 10 grams of Gelrite in 1 liter of media by boiling (add Gelrite slowly to gently boiling media) or autoclaving for 20 minutes on the liquid/fluid cycle.

7. Add 5 ml of 1M magnesium chloride / 0.3 M calcium nitrate solution (pre-made) after the Gelrite has completely dissolved (adding the Mg/Ca solution before the Gelrite has dissolved will prevent the Gelrite from completely dissolving, leading to plates that melt in the incubator)

8. Pour the plates in the laminar flow hood.