

## 926: ALKALIPHILIC THERMOCOCCUS MEDIUM

This recipe contains strain-specific modifications for *Thermococcus acidaminovorans* DSM 11906 \*

Final volume: 2000 ml

NaCl	27.70	g
MgSO <sub>4</sub> x 7 H <sub>2</sub> O	7.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	5.50	g
KCl	0.65	g
NaBr	0.10	g
NaHCO <sub>3</sub>	0.32	g
K <sub>2</sub> HPO <sub>4</sub>	1.00	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.05	mg
KI	15.00	mg
H <sub>3</sub> BO <sub>3</sub>	0.03	g
<b>Modified Wolin's mineral solution II</b>	20.00	ml
<b>Casamino acids</b>	<b>0.20</b>	<b>%</b>
Distilled water	2000.00	ml

1. Prepare the medium anaerobically under nitrogen. Do not adjust the pH.
2. Prepare separate anaerobic stock solutions of, Casamino acids(10%), Yeast extract (10%), and glycine (2M = 150 g/l). A 0.5M polysulphide solution is prepared by dissolving 12.0 g Na<sub>2</sub>S x 9 H<sub>2</sub>O in oxygen free water, followed by adding 1.6 g sulphur - the solution will be dark yellow.

\* To the sterile, anaerobic, mineral medium add 0.08 ml polysulphide/10 ml medium, casamino acids to a final concentration of 0.2%. There may be precipitation of material and the medium will turn pale yellow due to the addition of the polysulphide. The colour will disappear as the strain grows.

### Modified Wolin's mineral solution II (from medium 700)

Nitrilotriacetic acid	1.500	g
MgSO <sub>4</sub> x 7 H <sub>2</sub> O	3.000	g
MnSO <sub>4</sub> x H <sub>2</sub> O	0.500	g
NaCl	1.000	g
FeSO <sub>4</sub> x 7 H <sub>2</sub> O	0.100	g
CoSO <sub>4</sub> x 7 H <sub>2</sub> O	0.180	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.100	g
ZnSO <sub>4</sub> x 7 H <sub>2</sub> O	0.180	g
CuSO <sub>4</sub> x 5 H <sub>2</sub> O	0.010	g
AlK(SO <sub>4</sub> ) <sub>2</sub> x 12 H <sub>2</sub> O	0.020	g



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H <sub>3</sub> BO <sub>3</sub>	0.010	g
Na <sub>2</sub> MoO <sub>4</sub> x 2 H <sub>2</sub> O	0.010	g
NiCl <sub>2</sub> x 6 H <sub>2</sub> O	0.025	g
Na <sub>2</sub> SeO <sub>3</sub> x 5 H <sub>2</sub> O	0.300	mg
Distilled water	1000.000	ml

First dissolve nitrilotriacetic acid and adjust pH to 6.5 with KOH, then add minerals. Adjust final to pH 7.0 with KOH.