977. VULCANITHERMUS MEDIUM

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\begin{align*}
\text{NH}_4\text{Cl} & \quad 0.33 \quad \text{g} \\
\text{MgCl}_2 \times 6 \text{H}_2\text{O} & \quad 0.33 \quad \text{g} \\
\text{CaCl}_2 \times 2 \text{H}_2\text{O} & \quad 0.33 \quad \text{g} \\
\text{KCl} & \quad 0.33 \quad \text{g} \\
\text{KNO}_3 & \quad 0.33 \quad \text{g} \\
\text{NaCl} & \quad 25.00 \quad \text{g} \\
\text{PIPES (12mM)} & \quad 3.60 \quad \text{g} \\
\text{Yeast extract} & \quad 0.50 \quad \text{g} \\
\text{Tryptone} & \quad 1.00 \quad \text{g} \\
\text{Sucrose} & \quad 1.00 \quad \text{g} \\
\text{Vitamin (see medium 141)} & \quad 1.00 \quad \text{ml} \\
\text{Trace elements (see medium 141)} & \quad 1.00 \quad \text{ml} \\
\text{Distilled water} & \quad 1000.00 \quad \text{ml}
\end{align*}
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Prepare the medium anaerobically, under nitrogen, omitting the \(\text{CaCl}_2\), \(\text{MgCl}_2\), \(\text{KNO}_3\), tryptone, yeast extract, vitamins and sucrose. The pH should be 6.8. Dispense the medium into vessels suitable for anaerobic growth (Hungate tubes or serum bottles) under an atmosphere of nitrogen and autoclave. To the sterile, cooled medium add, from sterile stock solutions the \(\text{CaCl}_2\), \(\text{MgCl}_2\), \(\text{KNO}_3\), tryptone, yeast extract, vitamins and sucrose. The \(\text{CaCl}_2\), \(\text{MgCl}_2\), \(\text{KNO}_3\), tryptone, yeast extract, and sucrose stock solutions should be autoclaved, while the vitamin solution is sterile filtered. This strain grows anaerobically in the presence of nitrate, but aerobic growth has been reported provided the oxygen concentration does not exceed 16% (v/v). Under anaerobic conditions a redox agent, such as sulphide is not required.