

## 504. ANAEROBIC SEAWATER (SWM) MEDIUM

### Solution A:

KH <sub>2</sub> PO <sub>4</sub>	0.20	g
NH <sub>4</sub> Cl	0.25	g
NaCl	20.00	g
MgCl <sub>2</sub> x 6 H <sub>2</sub> O	3.00	g
KCl	0.50	g
CaCl <sub>2</sub> x 2 H <sub>2</sub> O	0.15	g
Na-resazurin solution (0.1% w/v)	0.50	ml
Distilled water	940.00	ml

### Solution B:

Trace element solution SL-10 (see medium 320)	1.00	ml
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### Solution C:

Vitamins solution (see medium 503)	1.00	ml
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### Solution D:

Selenite-tungstate solution (see medium 385)	1.00	ml
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### Solution E:

Na <sub>2</sub> CO <sub>3</sub>	1.50	g
Distilled water	30.00	ml

### Solution F:

Substrate solution (see below)

### Solution G:

Na <sub>2</sub> S x 9 H <sub>2</sub> O	0.30	g
Distilled water	10.00	ml

Sparge *solution A* with 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas mixture for 30 – 45 min to make it anoxic, distribute under same gas atmosphere into anoxic Hungate-type tubes or serum vials and autoclave. *Solutions B, D* and *G* are autoclaved separately under 100% N<sub>2</sub> gas. *Solutions C* and *F* are prepared under 100% N<sub>2</sub> gas and sterilized by filtration. *Solution E* is autoclaved under 80% N<sub>2</sub> and 20% CO<sub>2</sub> gas atmosphere. To complete the medium appropriate amounts of *solutions B* to *G* are added to the sterile *solution A* in the sequence as indicated. Adjust pH of complete medium to 7.2 - 7.4, if necessary.

**Substrates and additional instructions: see next page!**

For DSM 5848 and DSM 9537 use 2.50 g/l of Na<sub>2</sub>-succinate and 0.50 g/l yeast extract as substrates.

For DSM 6233 use 0.50 g/l of pyrogallol as the substrate. When growth appears feed once more with 0.50 g/l pyrogallol.

For DSM 12881, DSM 19012 and DSM 25728 use 2.00 g/l D-glucose and 0.50 g/l yeast extract as substrates.

For DSM 17953 and DSM 19335 use 2.50 g/l maltose and 2.00 g/l yeast extract as substrates.