

732a. DESULFUROMONAS MEDIUM (TCE)**Solution A:**

K ₂ HPO ₄	0.65	g
NaH ₂ PO ₄ x H ₂ O	0.17	g
Peptone (BD Bacto)	0.10	g
Na-acetate	0.46	g
Selenite-tungstate solution (see medium 385)	1.00	ml
Na-resazurin solution (0.1% w/v)	0.50	ml
Distilled water	900.00	ml

Solution B:

(NH ₄) ₂ CO ₃	0.27	g
NaHCO ₃	3.73	g
Distilled water	100.00	ml

Solution C:

CaCl ₂ x 2 H ₂ O	0.11	g
MgCl ₂ x 6 H ₂ O	0.10	g
Distilled water	10.00	ml

Solution D:

Trace elements solution (see medium 732)	1.00	ml
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Solution E:

Vitamin solution of medium 141	9.00	ml
Vitamin solution of medium 503	1.00	ml

Solution F:

Na ₂ S x 9 H ₂ O	0.30	g
Distilled water	10.00	ml

Solution G:

Hexadecane	13.50	ml
Tetrachloroethene	1.50	ml

Sparge *solution A* with 80% N₂ and 20% CO₂ gas mixture for 30 – 45 min to make it anoxic, then dispense under same gas atmosphere into anoxic serum vials (e.g., 9 ml in 50 ml bottles) and autoclave. *Solution B* is autoclaved separately under 80% N₂ and 20% CO₂ gas atmosphere. *Solutions C, D* and *F* are autoclave under 100% N₂ gas atmosphere.

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Solution E is prepared under 100% N₂ gas atmosphere and sterilized by filtration. Prepare *solution G* by filling 13.5 ml hexadecane into a 50 ml serum bottle, then sparge with 100% N₂ gas to make it anoxic and autoclave. Add 1.50 ml anoxic autoclaved tetrachloroethene to the sterile anoxic hexadecane solution by syringe. To complete the medium add appropriate amounts of *solutions B* to *F* to the sterile *solution A* in the sequence as indicated. The pH of the medium before inoculation should be at 7.2. Add solution G only after inoculation of the medium!

For DSM 13726 omit acetate from *solution A* and add 2.50 g/l Na-DL-lactate to the medium from a sterile anoxic stock solution prepared under 100% N₂ gas.