4: ASW

Final pH: 7.6 - 7.8
Final volume: 1000 ml

Mail sol. C_4

Tricine	0.50	g
Extra salts	3.75	ml
Vitamin solution	2.50	ml
SE1 (Soil Extract 1)	25.00	ml

1. Make up to 1 Litre with filtered natural seawater. Adjust pH 7.6 - 7.8 to with 1M NaOH or 1M HCl, prior to autoclaving. Autoclave at 15 psi for 15 minutes.

2. Alternatively, use: Distilled water to 1 Litre and “Ultramarine Synthetica” sea salts 33.60g

Extra salts

$NaNO_3$	30.00	g
Na_2HPO_4	1.20	g
K_2HPO_4	1.00	g

Vitamin solution

Biotin	0.0002	g
Calcium pantothenate	0.0200	g
Cyanocobalamine	0.0040	g
Folic acid	0.0004	g
Inositol	1.0000	g
Nicotinic acid	0.0200	g
Thiamine HCl	0.1000	g
Thymine	0.6000	g

SE1 (Soil Extract 1)

Soil (air-dried)	traces
Distilled water	traces

1. Site selection for a good soil is very important and for most purposes a soil from undisturbed deciduous woodland is best. Sites to avoid are those showing obvious signs of man's activity and particular care should be taken to avoid areas where fertilizers, crop sprays or other toxic chemicals may have been used.

2. A rich loam with good crumb structure should be sought. Stones, roots and larger

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invertebrates should be removed during an initial sieving through a 1 cm mesh. The sieved soil should be spread to air dry and handpicked for smaller invertebrates and roots. It should be turned periodically and picked over again. When dry sieve through a finer mesh (2-4 mm) and store in an airtight container away from light and heat.

3. Soil is prepared as above. Air-dried soil and twice its volume of supernatant distilled water are autoclaved together at 15 psi for 2 hours and left to cool. The supernatant is then decanted and then distributed to containers in volumes suitable for making up batches of media. The aliquots and their containers are autoclaved for an appropriate length of time (e.g. 1 litre or less for 15 minutes) and are then kept in a cool place (e.g. a refrigerator) until required.

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