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Aquil* Medium

Sunda, W.G., Price, N.M. and Morel, F.M.M. (2005)-see references below the recipe

Aquil* medium is a synthetic ocean water (SOW) that is enriched with macro- and micronutrients. Aquil* is a new version of the original Aquil medium (Morel *et al.* 1979, Price *et al.* 1989), and modifications are discussed Sunda et al. (2005). The medium, in all three versions, is designed for critical experimental work. To prepare properly, solutions are passed through a Chelex column to remove impurities. Details for column purification are thoroughly described (Sunda et al. 2005), and it is imperative that these details are followed when using the medium for critical experimental work. The recipes provided below are a brief summary of the components and their amounts. The first recipe is for preparation of the synthetic ocean water, the second recipe provides the macro- and micronutrients, and the third recipe describes the preparation of a vitamin solution.

To 600 mL of high quality dH_2O (e.g., MilliQ water), individually dissolve each of the anhydrous salts. To 300 mL of high quality dH_2O, individually dissolve each of the hydrous salts. Combine the two salt solutions. Add 1 mL of each major nutrient stock solution; add 1 mL of the trace metals solution and of the vitamin solution. Final salinity is 35 psu. The medium should be sterilized in a microwave oven or filter sterilized to avoid metal contamination from an autoclave.

Synthetic Ocean Water (SOW)

Component	Stock Solution	Quantity	Molar Concentration in Final Medium
Anhydrous Salts			
NaCl	---	24.5400 g	4.20×10^{-1} M
Na_2SO_4	---	4.0900 g	2.88×10^{-2} M
KCl	---	0.7000 g	9.39×10^{-3} M
$NaHCO_3$	---	0.2000 g	2.38×10^{-3} M
KBr	---	0.1000 g	8.40×10^{-4} M
H_3BO_3	---	0.0030 g	4.85×10^{-5} M
NaF	---	0.0030 g	7.15×10^{-5} M
Hydrous Salts			
$MgCl_2.6H_2O$	---	11.1000 g	5.46×10^{-2} M
$CaCl_2.2H_2O$	---	1.5400 g	1.05×10^{-2} M
$SrCl_2.6H_2O$	---	0.0170 g	6.38×10^{-5} M

Major Nutrients

The major nutrients may be prepared separately as stock solutions and 1 mL of each added to SOW to prepare 1 liter of medium. Alternatively, 10^{-3} of the stock solution may be mixed directly with the SOW salts.

Component	Stock Solution	Quantity	Molar Concentration in Final Medium
NaH_2PO_4 H_2O	1.38 g L^{-1} dH_2O	1 mL	1.00×10^{-5} M
$NaNO_3$	8.50 g L^{-1} dH_2O	1 mL	1.00×10^{-4} M
Na_2SiO_3 $9H_2O$	28.40 g L^{-1} dH_2O	1 mL	1.00×10^{-4} M

Metal/Metalloid Stock Solution

First, prepare the individual stock solutions using highest quality de-ionized water (e.g., MilliQ). Next, dissolve 29.2 g of EDTA (free acid, MW 292) in 950 mL of highest quality de-ionized water and add 1 mL of each stock solution. Bring the volume up to 1 liter, and use 1 mL of the final solution for preparing each liter of SOW.

Component	Stock Solution	Quantity	Molar Concentration in Final Medium
EDTA	---	29.200 g	1.00 x 10^{-4} M
$FeCl_3.6H_2O$	---	0.270 g	1.00 x 10^{-6} M
$ZnSO_4.7H_2O$	---	0.023 g	7.97 x 10^{-8} M
$MnCl_2.4H_2O$	---	0.0240 g	1.21 x 10^{-7} M
$CoCl_2.6H_2O$	---	0.0120 g	5.03 x 10^{-8} M
$Na_2MoO_4.2H_2O$	---	0.0242 g	1.00 x 10^{-7} M
$CuSO_4.5H_2O$	4.9 g L^{-1} dH_2O	1 mL	1.96 x 10^{-8} M
Na_2SeO_3	1.9 g L^{-1} dH_2O	1 mL	1.00 x 10^{-8} M

Mixed Vitamin Stock Solution

To prepare, first make separate stock solutions of cyanocobalamin and biotin by dissolving the indicated amounts into 1 liter of highest quality de-ionized water (e.g., MilliQ). To prepare the mixed vitamin stock solution, begin with 950 mL of high quality de-ionized water, add 1 mL of the cyanocobalamin stock solution, 1 mL of the biotin stock solution and 100 mg of thiamine. Bring the final volume to 1 liter with de-ionized water. After completely dissolved, filter sterilize the solution and dispense into small containers (e.g., 1-10 mL aliquots) and freeze. Use 1 mL of the mixed vitamin stock solution for each liter of Aquil* medium.

Component	Stock Solution	Quantity	Molar Concentration in Final Medium
Thiamine (Vit. B_1)	---	100 mg	2.97 x 10^{-7} M
Biotin (Vit. H)	5.0 g L^{-1} dH_2O	1 mL	2.25 x 10^{-9} M
cyanocobalamin (Vit.B_{12})	5.5 g L^{-1} dH_2O	1 mL	3.70 x 10^{-10} M

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Sunda, W.G., Price, N.M., and Morel, F.M.M. 2005. Trace metal ion buffers and their use in culture studies (Chapt. 4) pp. 35-63. In Andersen, R.A. (Ed.) *Algal Culturing Techniques*. Acad. Press/Elsevier, Amsterdam.