**Media Recipes**

½ Marine Agar Plates **(**makes 1 l**)**

1 l double distilled water

9 g Sodium Chloride (Sigma)

18 g Marine Broth 2216 (Difco)

15 g Bacto Agar (Difco)

L1 Algal Medium (makes 1 l)

\*\*This recipe is based on L1 Medium from Guillard and Hargraves (1993) [https://ncma.bigelow.org/node/83].

1. Add 800 ml filtered and autoclaved seawater to 1 L measuring cylinder.
2. Add the various components (stored in the 4°C fridge) according to table below in the laminar flow hood (measurements good for 1 l)

|  |  |  |  |
| --- | --- | --- | --- |
| **Component** | **Stock solution** | **Quantity** | **Molar concentration in final medium** |
| NaNO3 | 75.00 g l-1dH2O | 1 ml | 8.82 x 10-4 M |
| NaH2PO4.H2O | 5.00 g l-1dH2O | 1 ml | 3.62 x 10-5 M |
| Na2SiO3.9H2O | 30.00 g l-1dH2O | 1 ml | 1.06 x 10-4 M |
| Trace element solution | (see recipe on other L1 media protocol) | 1 ml | --- |
| Vitamin solution | (see recipe on other L1 media protocol) | 0.5 ml | --- |

1. Top up cylinder to 1 l with the seawater.
2. Use a 1 l filtering unit (Nalgene), filter into a sterile bottle (can re-use filters in single day by filtering into 1 l autoclaved Pyrex bottles).

*Notes:*

* Do all the addition of nutrient solutions in the lamina flow hood.
* All the components for the media should be aliquoted for 1 l of medium into a cryovial and stored in the fridge (this should all be done in the laminar flow hood). Label each tube with date, initials, name and volume.
* Precipitate in the glass bottles is normal – it is not biological, but is SiO2.
* To clean the algal culture tubes, collect all the lids into a beaker, cover with MilliQ water, covering the beaker with aluminium foil, then autoclave. The water will be tea colored after autoclaving. Repeat at least 4 times or until the water is clear.

References

Guillard RRL, Hargraves PE. 1993. *Stichochrysis immobilis* is a diatom, not a chrysophyte. Phycologia 32(3): 234-236.