L1+

(Modified from: Guillard R. R. L. and Hargraves P. E., 1993. *Stichochrysis immobilis* is a diatom, not a chrysophyte. *Phycol*. 32: 234-236.)

Filter 1L of natural sea water with GF/F (0.45 μm) and autoclave for 15 min at 120°C.

After cooling, add the following compounds with a sterile filter (0.2 µm)to complete the media:

Minerals	Stock Solutions (g/L)	Quantity (mL Stock/L Media)	Molarity (mM)
NaNO ₃	150	0.5	0.8
NaH ₂ PO ₄ · 2H ₂ O	6.9	0.725	0.03
Trace Element Solution	See recipe below	1	-
Vitamins3 Mix	See recipe below	1	-
NH ₄ Cl	26.7	0.3	0.15

For solid medium use 7g/L of agarose. Sterilize the agarose separately in 550 ml of milliQ water. In this case the mineral solution is filled up to 400 ml.

Trace Element Solution:

Trace metals	Stock Solution (g/L mQ)	Quantity used/L
Na ₂ -EDTA . 2H ₂ O	-	4.36 g
FeCl ₃ . 6H ₂ O	-	3.15 g
MnCl ₂ . 4H ₂ O	178.1	1 mL
ZnSO ₄ . 7H ₂ O	2.3	10 mL
CoCl ₂ . 6H ₂ O	1.19	10 mL
CuSO ₄ . 5H ₂ O	2.5	1 mL
Na ₂ MoO ₄ . 2H ₂ O	19.9	1 mL
$Na_2SeO_3.5H_2O$	2.63	1 mL
NiSO ₄ . 6H ₂ O	2.63	1 mL
V_2O_5	0.09	10 mL
K ₂ CrO ₄	1.94	1 mL

Vitamins 3 Mix:

Vitamins	Vitamins 3 mix (quantity/100mL)
Thiamine-HCl	10 mg
d-biotin*	100 μL (from a stock of 5 mg in 10mL)
VItamin B12	100 µL (from a stock of 5 mg in 10mL)

^{*}Dissolve first in 0.1 mL 2M NaOH. Then add 9.9 mL of mQ water.