F/2 medium:

For 400 mL of Autoclaved Artificial Sea Water:

- Trace Metals 0.4 mL
- Phosphate Stock 0.4 mL
- Vitamin Mix 200 µL
- $NaHCO_3$ (fresh) Stock 1.0 mL

adjust pH to 7.8-8.2

Stock Solutions: (hereafter,. molarities indicated are the final concentrations in F/2)

Trace Metal Stock Solution:

For 200 mL H_20 milliQ:

$FeCl_3$	0.13 g
EDTA	0.87 g
100 µL of $CuSO_4$ $5H_2O$	0.98 g/100 mL
100 µL of $ZnSO_4$ 7 H_2O	2,2 g /100 mL
100 µL of $CoCl_2$ $6H_2O$	1 g/100 mL
100 µL of $MnCl_2$ $4H_2O$	18 g/100 mL
100 µL of Na_2MoO_4 $2H_2O$	0.63 g/100 mL

Phosphate Stock Solution:

For 200 mL H_20 milliQ:

NaH_2PO_4 H_2O	1 g

$NaHCO_3$ (fresh) Stock:

in 10 mL H_20 milliQ:

$NaHCO_3$	0.42 g

- filter sterilise through 0.2 µm filter.

Vitamin Stock:

For 250 mL H_20 milliQ:

Biotin	250 µL
Vitamin B12	250 µL
Thiamine HCl	50.0 mg

Aliquot in 200 µL (or 400 µL) and freeze (keep at –20° C)

For Agar Plates:

- Prepare medium with 2X stock solution (no vitamins), but same amount of Artificial Sea Water (typically 200 mL) + (optionally add 20 mM HEPES, from filter sterilised solution)
- Prepare 3% w/v Noble Agar solution in double distilled water , and autoclave (typically 200 mL in a 500 mL bottle)
- Let the autoclaved agar solution cool to about 40° C (in a water bath)
- Add the 2X F/2 solution, mix and add vitamins
- Pour plates

Artificial Sea-Water:

For 1000 mL H_20 milliQ:

NaCl		23,38 g
$MgSO_4$ $7H_2O$		4,93 g
$CaCl_2$ $2H_2O$		1,47 g
KBr		0,20 g
KCl		0,75 g
$MgCl_2$ $6H_2O$	4,1 g	
Borate Stock (SW1)		1.0 mL
Nitrate Stock (SW2)		1.0 mL
Tris Stock (SW3)		5.0 mL

Adjust pH to about 8.0

Borate Stock SW1:

for 200 mL distilled wayer

H3BO3	2,46 g

Nitrate Stock SW2:

for 200 mL distilled wayer

$NaNO_3$	15 g

TRIS Stock SW3:

for 200 mL distilled wayer

TRIS hydrochloride,	7.29 g
TRIZMA base	2.71 g