Exp. lists for

Disentangling the interactions between microbial loop and grazing food web at the southern ECS

[Equipment]

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Item | Type | Quantity  for 1 set of exp | Quantity  for all XX sets | Extras |
| 20-L carboys |  | 4 |  | 1 |
| 20-L bucket |  | 1 |  | 0 |
| 2-L carboys | Exact volume is 2.41 L | 22 |  | 3 |
| 20 μm nylon filters | 2m x 2m | 2 |  | 0 |
| Silicone tubes covered with 20 μm mesh |  | 4 |  | 0 |
| Silicone tube covered with 50 μm mesh |  | 1 |  | 0 |
| 50μm Norpac net |  | 1 |  | 0 |
| Flowmeter |  | 1 |  | 0 |
| Peristaltic pump |  | 1 |  | 0 |
| Silicone tubes 2 #1 | For pump rotate | 4 |  | 0 |
| Silicone tubes 3 #2 | For pump to filter holder | 4 |  | 0 |
| Filter holder |  | 2 |  | 0 |
| Plastic joint 1 | For tube 2 to tube 3 | 1 for 1 pump set |  | 0 |
| Plastic joint 2 | For filter holder to tube 3 | 1 for 1 pump set |  | 0 |
| Parafilm | For sealing 2L carboys | 1 |  | 0 |
| 1.2 μm filter #3 |  | 2 |  | 0 |
| 0.2 μm filter #3 |  | 2 |  | 0 |
| Kimwipes Kimtech | 11 x 21cm | 1 |  | 0 |
| Tweezer | For arranging filters | 1 |  | 0 |
| Portable FlowCAM  with fluorescent trigger |  | 1 |  | 0 |
| 100L dark incubation tank |  | 1 |  | 0 |

#1 Silicone tube 2 needs to match the peristaltic pump

#2 Silicone tube 3 needs to match the filter holder

#3 The 1.2 and 0.2 μm filters need to match the filter holder

[Chemicals]

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Item | Type | Amount  for 1 set of exp | Amount  for all XX sets | Extras |
| 10% paraformaldehyde |  | 1.68 mL |  | 0 |
| 50% glutaraldehyde |  | 12 mL |  | 0 |
| 40 % formalin  10% formalin |  | 5 mL  20 mL |  | 0 |
| 75% Ethanol | For sterilization | 0.5L |  | 0 |

[Consumables]

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Item | Type | Quantity  for 1 set of exp | Amount  for all XX sets | Extras |
| 2 mL cryogenic tubes  (bacteria) |  | 3 (T0) +  36 (T12; trmt1-6 \* 2rep \* 3subsample) |  | 11 |
| 50 mL centrifuge tubes  (HNF) |  | 3 (T0) +  36 (T12; trmt6-11 \* 2rep \* 3subsample) |  | 11 |
| 100 mL plastic bottle  (mzp) |  | 1 (T0) +  6 (Tend; trmt1-2 \* 3rep) |  | 3 |

[Seawater volume]

65 L (at least) =

20 L (one 20-L carboy) 0.2μm-filtered water (treatment 1-6) +

25 L (1.5 20-L carboy) 20μm-filtered water (treatment 2-10) +

5 L water with microzooplankton (treatment 11) +

20 L 20μm-filtered water (temporarily keep microzooplankton)

**(Exp III) Microzooplankton (mainly nauplii and copepodites) growth and production**

Secondary production of microplankton group *i* (*SPi*) is defined as

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where *GRi* is the mass-specific growth rate and *Bi* is the biomass of group *i*. We will incubate two microzooplankton size classes: 50-80 μm (nauplii) and 100-150 μm (copepodites).

20 L cubitainers will be filled with 18 L 50 μm-filtered incubation water (at 10m depth) with plankton < 50 μm. Nauplii and copepodites will be collected by 10-min vertical tow of 50 and100 μm plankton nets at 10 m depth and gently resuspended in a bucket of 20 L 50 μm-filtered incubation water. Fill the remaining space of cubitainers with nauplii/copepodites grazer by reverse filtering the seawater with nauplii/copepodites through 80/150 μm sieves. Nauplii and copepodites will be incubated for 24 and 48 hrs, respectively. Microzooplankton community before and after incubation will be preserved in 5% formalin for analysis.

[Equipment]

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Item | Type | Quantity  for 1 set of exp | Quantity  for all XX sets | Extras |
| 20 L cubitainers |  | 6  (2trmt\*3reps) |  | 6  (1 set) |
| dark incubation tank |  | 2 |  | 0 |
| 50 μm Norpac net |  | > 15  (in case it breaks) |  | 0 |
| 100 μm Norpac net |  | > 5  (in case it breaks) |  | 0 |
| Flowmeter for Norpac |  | 2 |  | 0 |
| Cod End for Norpac |  | 5 |  | 0 |
| 80 μm sieves for reverse filtration |  | 2 |  | 0 |
| 150 μm sieves for reverse filtration |  | 2 |  | 0 |
| 50 μm nylon filter |  | 2 |  | 0 |
| 50 um hand-made sieve |  | 2 |  | 0 |
| 20 L plastic buckets |  | 2 |  | 0 |
| 100 mL plastic bottles |  | 8 |  | 16 |
| Cleaning Sponge with Scouring Pad |  | 2 |  | 0 |
| Pressure sprayer  (https://tg.pe/rJu) |  | 2 |  | 0 |
| Alcohol thermometer |  | 2 |  | 0 |
| Wash bottle |  | 3 |  | 0 |

[Chemicals]

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Item | Type | Amount  for 1 set of exp | Amount  for all XX sets | Extras |
| formalin | 40 % or  10% | 40 mL  or  160 mL |  | 0 |

[Seawater volume]

160 L (at least) =

120L 50μm-filtered water (cubitainers) +

40L 50μm-filtered water (buckets)

**[I haven’t modified any of the following]**

**\* Time for net tows per station**

|  |  |  |  |
| --- | --- | --- | --- |
| Sampling depth | Mesh-size | Spend-time (minutes) | Experiment |
| 10-m vertical tow | 50 μm Norpac with a flowmeter | 15 (The net will be set to 10 m depth and allowed to drift with the ship for 10 minutes) | I |
| 10-m vertical tow | 50 μm Norpac\*2 and 100 μm Norpac attached on ring | 15 (The net will be set to 10 m depth and allowed to drift with the ship for 10 minutes) | II |
| From bottom to surface oblique tow | 50 μm Norpac with Flowmeter | 15-50 min, depending in the bottom depth | II |

**\* Go-Flo bottle water collection per station**

|  |  |  |
| --- | --- | --- |
| Sampling depth | # of Go-Flo (20L) | Experiment |
| 10-m | 3 | I |
| 10-m | 8 | II |