*Sampling*

Zooplankton metabolic contributions to active flux were calculated using abundance and dry mass analysis of the MOCNESS-1 on the RRS *James Cook*. Each net tow sample was split using a Folsom plankton splitter and processed using protocols described in Steinberg et al. (2008). Half of the sample was size-fractionated using nested sieves (200, 500, 1000, 2000, and 5000 µm), rinsed onto pre-weighed 0.2 mm Nitex mesh filters, and frozen at -20oC for biomass analysis. The other half sample was further split for additional analyses, with a portion preserved in sodium borate-buffered 4% formaldehyde for Zooscan analysis. For the May 17th the formaldehyde sample was size-fractionated using the same nested sieves prior to preservation. There were abundant large pteropods (>1.5 cm) in the MOCNESS sampling. These individuals were removed from the whole net sample, individuals enumerated, photographed, and preserved as a separate size fraction for dry mass analysis.

*Dry Mass*

Dry mass for zooplankton were measured on a Sartorius BP211D or Mettler AE 160 balance. First samples were thawed on paper towels to remove excess water (~ 20 minutes), then dried for at least 24 h at 60oC and re-weighed. Dry biomass of each size fraction within a net (mg m-3) was determined by dividing the biomass by the seawater volume filtered through the net during sampling. The average dry mass of the pteropods was 105.3 mg.

*Zooscan of abundance*

To estimate mesozooplankton abundance and average size, the formalin preserved samples from May 17 were imaged with a ZooSCAN version 3 at 2400 dpi as described in Maas et al. (2021). Briefly, at least 1500 particles per size fraction were scanned after subsampling using a Motoda splitter (Motoda, 1959). Raw images were processed in ZooProcess (Gorsky et al., 2010, Vandromme et al., 2012), then uploaded to EcoTaxa (https://ecotaxa.obs-vlfr.fr/; Picheral et al., 2017) for machine assisted identification. The entirety of the May 17 scan was manually validated for taxonomy. This dataset provides the abundance of zooplankton from each size fraction from each depth interval after correcting for fraction imaged and volume filtered. This average dry mass, which varied day vs. night and between depths (Table G1), was used to calculate the average metabolic contribution of a representative zooplankter within each size fraction using the allometric equations detailed below.

Physiological rates for both the pteropods and the general mesozooplankton were calculated as:

where DM is the estimated average dry mass (mg) of an individual within a size fraction, RQ is 0.87 for copepods (Mayzaud et al. 2005), and 0.536 allows conversion from µL CO2 to µg C h-1. Temperature (TempC, °C) is the average temperature measured by the MOCNESS in the 200-1000 m interval and corrects the rates the average temperature below the flux boundaries (11.1°C). The remaining coefficients for the respiration equation are from Ikeda et al. (2001) and were chosen as the best predictive equation based on measured metabolic rates, while the equation for DOC comes directly from measured regressions to zooplankton size (Maas et al. 2021). POC was estimated as 31% of respiratory CO2 (Schnetzer and Steinberg 2002, Steinberg et al. 2012).

Once the rate for a representative organism for a particular net and fraction was computed, this was scaled to the community by multiplying by the estimated average size (Table G1) divided by the measured dry mass of that net (which equates to the abundance of individuals in a net for that tow) to calculate of hourly production of waste for the zooplankton community (mgC m-3 h-1). This protocol was required because the abundance of individuals was only fully enumerated for the May 17th tows.

To calculate total active flux below a specific boundary, all of the night time respiration of mesozooplankton and pteropods above that boundary were summed and then subtracted from the daytime respiration of zooplankton above the boundary. This assumes that the physiological processes of these absent organisms were performed below the flux boundary during the day. This hourly rate of waste production was multiplies by the day length (15 h during the cruise) to estimate the total daily active flux of the mesozooplankton community at each flux boundary.

Table SG1: Dry mass (mg) of an average mesozooplankton individual from each size fraction of the daytime or nighttime tow from May 17th, 2021. The large pteropod average mass was estimated to be 105.3 mg at all depths and times.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| D/N | depth | 200 (mg) | 500 (mg) | 1000 (mg) | 2000 (mg) | 5000 (mg) |
| Day | 1000-750 | 0.0015 | 0.0142 | 0.0561 | 0.5323 | 1.0815 |
| Day | 750-500 | 0.0032 | 0.0075 | 0.0384 | 0.1451 | 0.1939 |
| Day | 500-400 | 0.0014 | 0.0067 | 0.0555 | 0.1252 | 0.4306 |
| Day | 400-300 | 0.0016 | 0.0069 | 0.0404 | 0.0584 | 0.2641 |
| Day | 300-200 | 0.0015 | 0.0091 | 0.0723 | 0.2104 | 0.1885 |
| Day | 200-150 | 0.0016 | 0.0074 | 0.0309 | 0.1184 | 0.1868 |
| Day | 150-100 | 0.0017 | 0.0081 | 0.0243 | 0.2397 | 0.0717 |
| Day | 100-50 | 0.0011 | 0.0021 | 0.0034 | 0.0118 | 0.4692 |
| Day | 50-0 | 0.0015 | 0.0031 | 0.0135 | 0.1075 | 0.4995 |
| Night | 1000-750 | 0.0013 | 0.0109 | 0.0471 | 0.1298 | 1.2558 |
| Night | 750-500 | 0.0018 | 0.0072 | 0.0384 | 0.2897 | 0.4097 |
| Night | 500-400 | 0.0013 | 0.0056 | 0.0374 | 0.1973 | 0.1358 |
| Night | 400-300 | 0.0014 | 0.0047 | 0.0286 | 0.0771 | 0.1351 |
| Night | 300-200 | 0.0018 | 0.0052 | 0.0123 | 0.0834 | 0.4262 |
| Night | 200-150 | 0.0015 | 0.0053 | 0.0268 | 0.0853 | 0.4215 |
| Night | 150-100 | 0.0014 | 0.0043 | 0.0182 | 0.0816 | 0.2749 |
| Night | 100-50 | 0.0012 | 0.0037 | 0.0280 | 0.1042 | 0.3633 |
| Night | 50-0 | 0.0014 | 0.0031 | 0.0049 | 0.0190 | 0.1863 |

Table SG2: Flux contributions by the large pteropod and mesozooplankton community to active flux (mgC m-2 d-1) below specified flux boundaries during the three MOCNESS tow periods.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Flux boundary | Date | Respiration  (CO2) | Excretion (DOC) | Egestion  (POC) | Total C |
| 500 | May 11 | 13.66 | 14.50 | 4.24 | 32.40 |
|  | May 17 | 20.04 | 23.10 | 6.21 | 49.35 |
|  | May 26 | 10.56 | 16.80 | 3.27 | 30.64 |
| 200 | May 11 | 23.59 | 20.30 | 7.31 | 51.21 |
|  | May 17 | 31.71 | 27.87 | 9.83 | 69.40 |
|  | May 26 | 33.18 | 28.75 | 10.28 | 72.21 |