

## Comparison of two multiple plankton samplers: MOCNESS and Multinet Mammoth

Espen Strand \*, Cecilie Broms, Espen Bagøien, Tor Knutsen, Hilde Arnesen, Bahar Mozfar

Institute of Marine Research, Bergen, Norway

### Abstract

To ensure an optimal continuation of a long time series of zooplankton monitoring surveys, two types of equipment for depth-stratified mesozooplankton sampling were compared. The Institute of Marine Research (Norway) has applied the MOCNESS with good results since 1985, but recent events have made it necessary to change to the Multinet Mammoth. During a cruise in March 2019, both sampling devices were calibrated before 17 paired deployments of the 2 gears were undertaken. During each deployment, three nets and depth-strata covering ~ 425–200, 200–100, and 100–0 m were sampled. All samples were size-fractionated or taxonomically fractionated into 10 different biomass categories. The results revealed no significant differences between the two gears when comparing total depth-integrated biomass ( $2.46 \pm 0.36$  vs.  $2.61 \pm 0.59$  gDW m<sup>-2</sup>) or depth-integrated biomass of any specific biomass category. Running paired *t*-tests separately for all combinations of biomass categories and nets, the differences were only significant for zooplankton biomasses in the 180–1000 μm size fraction and only for Net 2. Possible reasons for this result are discussed in the paper. Gears produced similar catches whether sampling during day or night. We conclude that the MOCNESS and Multinet Mammoth in this study provided comparable results regarding abundances of various zooplankton categories.

Time-series analysis of year-class strength laid the foundation of Hjort's (1914) description of population dynamics in fisheries, and most biologists have heard the tale of hares and lynxes and their predator–prey relationship based on over 100 years of fur return data from trappers (Elton and Nicholson 1942; Maclulich 1936). In plankton ecology, the continuous plankton recorder survey has been ongoing since 1931 and continues to be of great importance (Beaugrand et al. 2009; Reid et al. 2003). In a general perspective, a time-series scientific value will increase with several factors such as timespan, sampling continuity, and sampling frequency, as well as consistency in sampling procedures and sampling gear. Nonetheless, sometimes changes to the established continuity of a time series are unavoidable. It is then of paramount

\*Correspondence: [espen.strand@hi.no](mailto:espen.strand@hi.no)

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**Author Contribution Statement:** Espen Strand (conception, data acquisition, analysis, drafting of manuscript), Cecilie Broms (conception, data acquisition, drafting of manuscript), Espen Bagøien (conception, drafting of manuscript), Tor Knutsen (conception, drafting of manuscript), Hilde Arnesen (data acquisition, analysis), Bahar Mozfar (data acquisition, analysis)

importance to understand how such a change in method and/or effort will affect the time series in general.

The history of plankton sampling equipment goes back to at least Charles Darwin onboard the *Beagle*, where he in 1832 in his diary (Darwin 1988) draws and describes his plankton net in action outside the Cape Verde islands: “I proved today the utility of a contrivance which will afford me many hours of amusement & work—it is a bag four feet deep, made of bunting, & attached to [a] semicircular bow this by lines is kept upright, & dragged behind the vessel—this evening it brought up a mass of small animals, & tomorrow I look forward to a greater harvest.”

Since then there has, naturally, been a strong motivation to develop better and more precise plankton samplers (Wiebe and Benfield 2003). At the Institute of Marine Research (IMR), Bergen, Norway, the MOCNESS (Wiebe et al. 1976, 1985) has been used as the standard equipment for stratified mesozooplankton sampling from 1984 until 2018. During that time, a total of 9 different research vessels on 319 distinct cruises have performed a total of 3742 MOCNESS deployments and collected a total of 23,438 MOCNESS net samples. In 2018, it was decided that the MOCNESS was to be replaced with the Multinet Mammoth (Hydro-Bios 2020) due to the challenge of maintaining the MOCNESS hardware, electronics and software for high capacity and stable year-round operations. Both gears have a 1 m<sup>2</sup> mouth opening with nine available nets (180 μm mesh)

that can be opened and closed in sequence at predetermined or manually targeted depths. Although both gears appear similar in terms of opening area and number of nets, their designs are quite different in terms of net open/close mechanism as well as how they move through the water. To ensure that the transition from MOCNESS to Multinet Mammoth would not compromise the long and extensive time series of depth-stratified zooplankton sampling at IMR, it was decided to run a controlled test of both equipment during a 3-d cruise in spring 2019 in a fjord south of Bergen, Norway.

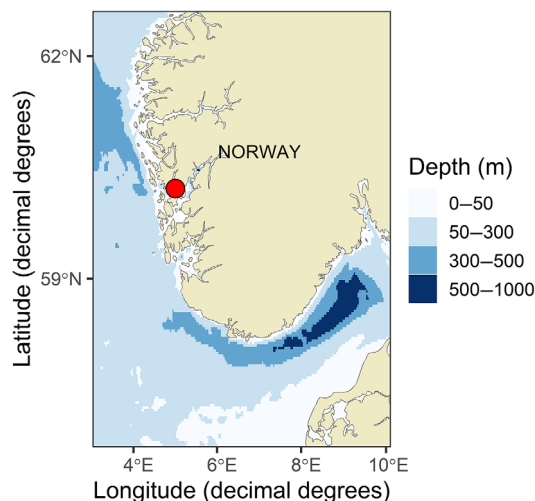
To our knowledge, a direct comparison of MOCNESS and Multinet has only once before been published (Skjoldal et al. 2013). However, that study compared the Multinet Midi (0.25 m<sup>2</sup> opening and with the standard 200  $\mu$ m mesh) with a range of other plankton samplers, including the 1 m<sup>2</sup> MOCNESS with 180  $\mu$ m mesh size used in the present study.

This paper focuses on comparing the sampling results of these two multiple plankton samplers by measuring the surface integrated biomass (gDW m<sup>-2</sup>) or the biomass density (mgDW m<sup>-3</sup>) of various sizes of mesozooplankton as well as selected groups of larger macrozooplankton. Our null hypothesis is that these two sampling gears collect the same biomass of all species and groups, at different depths, and during day and night.

## Materials and procedures

### Study area

All calibration and sampling were made during the period 24–26 March 2019 from the research vessel G.O. Sars in Bjørnafjorden (60.136°N, 5.616°E) on the west coast of Norway, just south of Bergen (Fig. 1). The rationale behind choosing this fjord for the comparison experiment was as follows. First, it is sufficiently deep to allow for stratified



**Fig. 1.** Sampling location. Bjørnafjorden, Norway (60.136°N, 5.616°E).

sampling. Second, the fjord system is well studied, and it was expected to find mesozooplankton of various sizes as well as larger macrozooplankton and micronekton. Last, it provided enough shelter to work continuously without needing to consider weather conditions.

### Calibration

The MOCNESS operating software uses a calibration factor in the calculation of filtered sampling volume, and this is based on measuring how many meters the MOCNESS moves through the water per revolution of the flowmeter. For the Multinet Mammoth, a fixed calibration factor is set by the manufacturer and used for the volume calculation in the software (i.e., precalibrated).

For the MOCNESS, the standard calibration procedure was followed (Anonymous 1999; Dr. P.H. Wiebe, Woods Hole pers. comm.) by towing the MOCNESS in a straight line with Net 1 open for approximately 1 nautical mile (nm) while recording the number of flowmeter revolutions. Since the distance is measured as the great-circle distance (shortest distance on a sphere) between start and stop position determined by satellite (GPS), any water currents present will affect the result by reducing or increasing the relative speed of the equipment through the water during the 1 nm tow and thus the number of revolutions logged by the flowmeter. To account for this effect, the calibration procedure is repeated, only in the direct opposite direction of the first tow. The average of the two tows will make the basis for the calibration (Table 1).

Since the Multinet Mammoth flow meters come precalibrated from the manufacturer, there is to our knowledge no available calibration manual. However, it was decided to run the Multinet flow meter calibration in a similar way to the MOCNESS calibration to compare the estimated volumes of the two gears. Following a suggestion from Dr. R.D.M. Nash (Bergen pers. comm.) the Multinet was lowered to 30 m depth and towed at constant depth in a straight line for 1.5 nautical miles. Contrary to the described MOCNESS calibration, a new Multinet net was opened every 0.5 nautical mile, thus returning 3 separate measurements from within the standard software supplied by Hydro-Bios for the Multinet along the 1.5 nm transect. As with the MOCNESS, the procedure was immediately repeated in the opposite direction (a reciprocal tow). The estimated average volume filtered by the Multinet was 2101 m<sup>3</sup> nm<sup>-1</sup> compared to 2060 m<sup>3</sup> nm<sup>-1</sup> in the MOCNESS (Table 1), effectively meaning a ~ 2% difference in sampled volume. IMR has several Multinet Mammoth systems, basically one unit for each of its larger sea-going vessels. The Multinet Mammoth unit used on board R/V G.O. Sars for the current experiment, had the identification, IdentNo.: 4618.

### Sampling procedure

The sampling protocol was designed so that paired comparisons of the two gears could be made in the following analysis. A “pair” is hereafter defined as one deployment of the MOCNESS and one with the Multinet Mammoth that

**Table 1.** Results from the calibration prior to sampling. D1 and D2 refers to the two opposite directions towed during calibration. N1–N3 refers to net in use.

	Start (h)	Stop (h)	Distance (m)	Speed (knot)	Volume (m <sup>3</sup> )	Flow counts	Calibration factor (m count <sup>-1</sup> )	Volume (m <sup>3</sup> nm <sup>-1</sup> )
MOCNESS D1–N1	16 : 03 : 45	16 : 37 : 56	1883	1.76	2143	331	5.7	2108
MOCNESS D2–N1	17 : 03 : 22	17 : 37 : 25	1881	1.76	2043	297	6.3	2011
MOCNESS mean			1882		2093	314	6.0	2060
Mammoth D1–N1	13 : 07 : 49	13 : 25 : 01	961	1.74	1284			2475
Mammoth D1–N2	13 : 25 : 02	13 : 42 : 03	935	1.76	1303			2582
Mammoth D1–N3	13 : 42 : 04	13 : 59 : 08	934	1.76	1196			2372
Mammoth D2–N5	14 : 13 : 53	14 : 32 : 53	1056	1.58	1043			1829
Mammoth D2–N6	14 : 32 : 54	14 : 50 : 02	936	1.75	894			1769
Mammoth D2–N7	14 : 50 : 03	15 : 07 : 04	939	1.76	813			1604
Mammoth mean			960		1089			2101

Values in italics denote basic mean values.

are intended to be compared in the following statistical analysis. To limit the variation within a pair, all pairs were towed from the same starting position and in the same direction with as little time as possible in between deployments. Each deployment of either MOCNESS or Multinet were following the same procedures: By use of remote control, Net 1 was opened at the deepest point on the oblique trajectory (~ around 425 m, see Table 2) and hauled in with a winch speed of ~0.5 ms<sup>-1</sup>, resulting in an upwards movement of the equipment of approximately 0.4 ms<sup>-1</sup>. At 200 m depth, Net 2 was opened and thereby simultaneously closing Net 1. At 100 m depth Net 3 was opened and upon reaching the

surface, Net 3 was closed by opening Net 4. To ensure that the two gears were used in varying order within the pairs, and thus avoiding that a specific gear was predominantly used first or last with each pair, the protocol seen in Table 2 where one gear was deployed two times before switching gear was followed. Only at the start and end of the sampling procedure, marking the start of PAIR1 and the end of pair PAIR17, was a gear deployed only once before being switched. This was also an advantage in order to limit the amount of time spent disconnecting one gear and connecting the other to the towing cable and other electronic connections.

**Table 2.** Sampling design and overview of key sampling events and results.

Pair	Day (Mar 2019)	MOCNESS			Multinet Mammoth		
		Start time (UTC)	Max depth (m)	Total volume (m <sup>3</sup> )	Start time (UTC)	Max depth (m)	Total volume (m <sup>3</sup> )
1	25.	00 : 57	418	1230	02 : 21	421	1034
2	25.	04 : 30	414	1191	03 : 23	427	1095
3	25.	05 : 34	423	1218	07 : 08	426	1120
4	25.	09 : 18	423	1180	08 : 08	426	1117
5	25.	10 : 25	425	1139	11 : 38	429	1141
6	25.	13 : 44	421	1200	12 : 39	428	1115
7	25.	14 : 48	423	1176	15 : 51	429	1061
8	25.	18 : 16	428	1126	17 : 05	430	1118
9	25.	19 : 18	427	1175	20 : 21	428	1090
10	25.	23 : 21	421	1198	21 : 19	428	1118
11	26.	00 : 32	422	1193	01 : 39	427	1043
12	26.	03 : 45	420	1166	02 : 40	428	1108
13	26.	04 : 46	420	1195	06 : 00	426	1113
14	26.	08 : 25	424	1195	07 : 09	427	1128
15	26.	09 : 26	422	1218	10 : 37	428	1127
16	26.	13 : 21	423	1188	11 : 44	429	1129
17	26.	14 : 27	424	1171	15 : 27	429	1127

Based on the bathymetry at the sampling location, the lower sampling depth was set to 425 m, or as close to the bottom as safely possible. The rationale behind covering the entire water column down to as close to the bottom as possible was to lessen, if present, the impact of diel vertical migration that would result in animals migrating above or below a shallower sampling depth depending on day/night habitat preferences. Choosing to sample from the bottom to the surface should, in theory, mean that depth-integrated data (all three nets combined) would be easier to compare independent of day and night due to vertical migration. The reason for only using three of the eight available nets was simply to reduce the workload in the lab on board so that a maximum number of comparison pairs could be made.

### Sample treatment

Once the MOCNESS or Multinet was retrieved, the vessel turned around and moved back to its starting position. Meanwhile, the three biological samples were taken into the lab and worked up in accordance with standard IMR zooplankton procedures (Melle et al. 2004).

**Table 3.** Overview and description of biomass categories.

Biomass categories	Category description
Amphipods	Biomass of Amphipods (Amphipoda) retained on the 2000- $\mu\text{m}$ sieve
<i>Boreomysis</i>	Biomass of <i>Boreomysis</i> sp. (Mysida) retained on the 2000- $\mu\text{m}$ sieve
<i>Calanus hyperboreus</i>	Biomass of <i>Calanus hyperboreus</i> retained on the 2000- $\mu\text{m}$ sieve
Chaetognatha	Biomass of Chaetognaths retained on the 2000- $\mu\text{m}$ sieve
<i>Paraeuchaeta</i>	Biomass of <i>Paraeuchaeta</i> spp. retained on the 2000- $\mu\text{m}$ sieve
Shrimps	Biomass of shrimps (Caridea) retained on the 2000- $\mu\text{m}$ sieve
Fish	Biomass of fish (Teleostei) retained on the 2000- $\mu\text{m}$ sieve
Krill	Biomass of krill (Euphausiids) retained on the 2000- $\mu\text{m}$ sieve
GT2000_indet	Remaining biomass left on the 2000- $\mu\text{m}$ sieve after species/groups above had been removed
BT180.1000	Biomass that passed through the 1000- $\mu\text{m}$ sieve but was retained on 180 $\mu\text{m}$ .
BT1000.2000	Biomass that passed through the 2000- $\mu\text{m}$ sieve but was retained on 1000 $\mu\text{m}$ .
BT180.2000	Summed biomass of BT180.1000 + BT1000.2000
tot_GT2000	Summed biomass of everything retained on the 2000- $\mu\text{m}$ sieve
tot_biom	Summed biomass of everything in the net

All samples were treated the same way. First, the sample was carefully transferred to a Motoda plankton splitter (Motoda 1959) and by eye inspected to determine if there were any large non-numerous specimens present (typically mesopelagic fish or pelagic shrimps). If present, such individuals were removed from the sample, speciated, length-measured, and slightly rinsed in freshwater and placed on a preweighed aluminum dish to be dried and subsequently weighed. It was also noted that the specimen(s) were removed from the total sample. Next, the sample was split into two equal halves and one part transferred to a 100-mL plastic bottle using seawater and preserved by adding 10 mL of concentrated formalin, resulting in a  $\sim 4\%$  borate-buffered formalin sample. The other half of the sample was size-fractionated by successive sieving through screens with mesh-sizes 2000 and 1000  $\mu\text{m}$  before finally collecting the remaining part of the sample on screen with mesh-size 180  $\mu\text{m}$ . The biological material retained on the 2000- $\mu\text{m}$  sieve was gently sprinkled with freshwater to remove excess salt water before the individuals were carefully removed, counted and sorted into the following categories before being placed on aluminum dishes and dried: Chaetognaths, *Paraeuchaeta* spp., *Calanus hyperboreus*, the mysid *Boreomysis* spp., Krill, Fish, Amphipods, and Shrimps. In the latter four categories, individuals were also speciated (if possible) and their lengths measured. All individuals not belonging to the above-mentioned groups were placed on a separate dish labeled “GT2000\_indet.” Amphipods were only found in 2 of the 34 gear deployments and will thus not be explicitly presented in the results. Still, their weight will be included in the total biomass larger than 2000  $\mu\text{m}$  (tot\_GT2000) and total biomass overall (tot\_biom). See Table 3 for description of variables.

The part of the sample not retained by the 2000- $\mu\text{m}$  screen, was then sieved on 1000  $\mu\text{m}$ , and passing material was collected on a 180- $\mu\text{m}$  screen (which is the same mesh-size as used on the sampling nets of the MOCNESS and Multinet). The retained biological material on each sieve was carefully rinsed in freshwater before transferred to aluminum dishes labeled “BT1000.2000” and “BT180.1000,” respectively. All dishes had individual numbering and had been preweighed on shore with a Sartorius Quintix 224-1CEU weight with an accuracy of 0.0001 g. After size-fractionation and sorting of the samples, all aluminum dishes were placed in a drying cabinet at 60 °C for at least 12 h or until dry.

Following the cruise, all aluminum dishes with biological material were once more dried overnight in the lab to ensure stable dry weight prior to weighing. The dishes were weighed on the same scale that was used to preweigh the empty dishes.

### Data management and statistical analysis

All postprocessing and visualization of data were done using R (R Core Team, 2021) and the R library package Tidyverse (Wickham et al. 2019).

Measured biomass (gDW) on all trays was divided by corresponding sampled volume ( $\text{m}^3$ ) to obtain average biomass density ( $\text{gDW m}^{-3}$ ) for each biomass category in each net.

Surface integrated values of total biomass ( $B$ ;  $\text{gDW m}^{-2}$ ) were calculated using Eq. 1 where  $\rho$  is the density ( $\text{gDW m}^{-3}$ ) of biomass component  $i$  (11 in total) in net  $j$  (3 in total) during deployment  $d$ .  $L$  and  $U$  are lower and upper depths of net  $j$  during deployment  $d$ .

$$B_d = \sum_{i=1}^{11} \left( \sum_{j=1}^3 \left( \rho_{i,j,d} * (L_{j,d} - U_{j,d}) \right) \right). \quad (1)$$

When calculating surface integrated biomass of specific biomass categories, the outer summation loop is omitted.

All statistical analyses were performed using paired, two-sided  $t$ -tests. Paired  $t$ -tests are generally considered more powerful than unpaired  $t$ -tests as between-pair variation is blocked out, hence making the test more sensitive. In our case, if we had tested the gears sequentially and not in an alternating pairwise manner, this could have led to confounding by the possible inclusion of effects of changes in currents, light, predation, or growth.

In the cases where both biomass categories (10) and nets (3) are tested simultaneously (i.e., 30 or more  $t$ -tests), the BH method (Benjamini and Hochberg 1995) for correction for multiple testing is added (Kassambara 2021). This method controls the false discovery rate using sequential modified Bonferroni correction for multiple hypothesis testing.

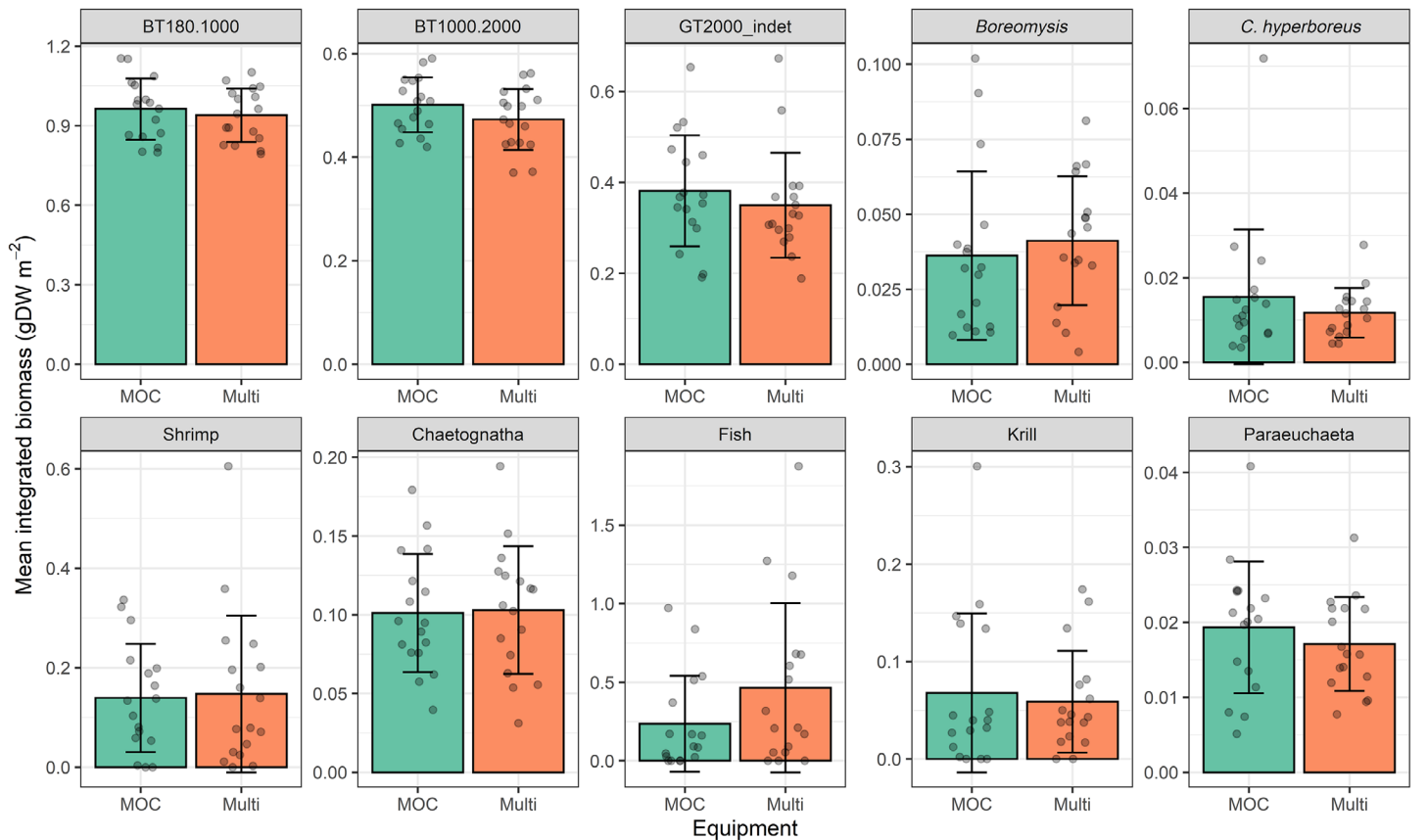
## Assessment

### Total biomass vs. equipment

First, the data were aggregated to calculate the total integrated surface biomass ( $\text{gDW m}^{-2}$ ). By calculating the mean and standard deviation for the 17 deployments of each equipment, the data are in its most aggregated form. The mean surface integrated total biomass of the two equipment showed no significant difference ( $p = 0.33$ , two-sided paired  $t$ -test) with a mean  $\pm$  standard deviation of the MOCNESS of  $2.46 \pm 0.36 \text{ gDW m}^{-2}$  and the Multinet  $2.61 \pm 0.59 \text{ gDW m}^{-2}$  (both  $n = 17$ ).

### Biomass of different components vs. equipment

With no significant differences in catch efficiencies revealed by looking at the total surface integrated biomass, the next step was to look at the same, but for each biomass component instead of total biomass (Fig. 2).



**Fig. 2.** Mean ( $n = 17$ ) of the surface integrated biomass for each biomass component. Bars show mean with  $\pm$  SD error bars. Gray dots show all data. Note different range of the y-axis. All  $p$ -values  $\geq 0.12$  in paired, two-way  $t$ -test.

Still there are no significant differences between equipment ( $p \geq 0.12$  for all categories), but some of the biomass components show a very large standard deviation, especially the category Fish. The fish component consisted mainly of the mesopelagic fish *Benthoosema glaciale* (23 and 28 individuals in the MOCNESS and Multinet, respectively) and some *Maurolanicus muelleri* (6 and 3 individuals in the MOCNESS and Multinet, respectively) in addition to 2 fish larvae. In addition, the number of *B. glaciale* caught in different pairs ranged from 0 to 5 in both equipment with an average catch of 1.4 and 1.6 individuals per deployment in the MOCNESS and Multinet, respectively. Combining data from all deployments (both gears), we estimate an individual density of *B. glaciale* of  $0.0013 \text{ ind.m}^{-3}$ .

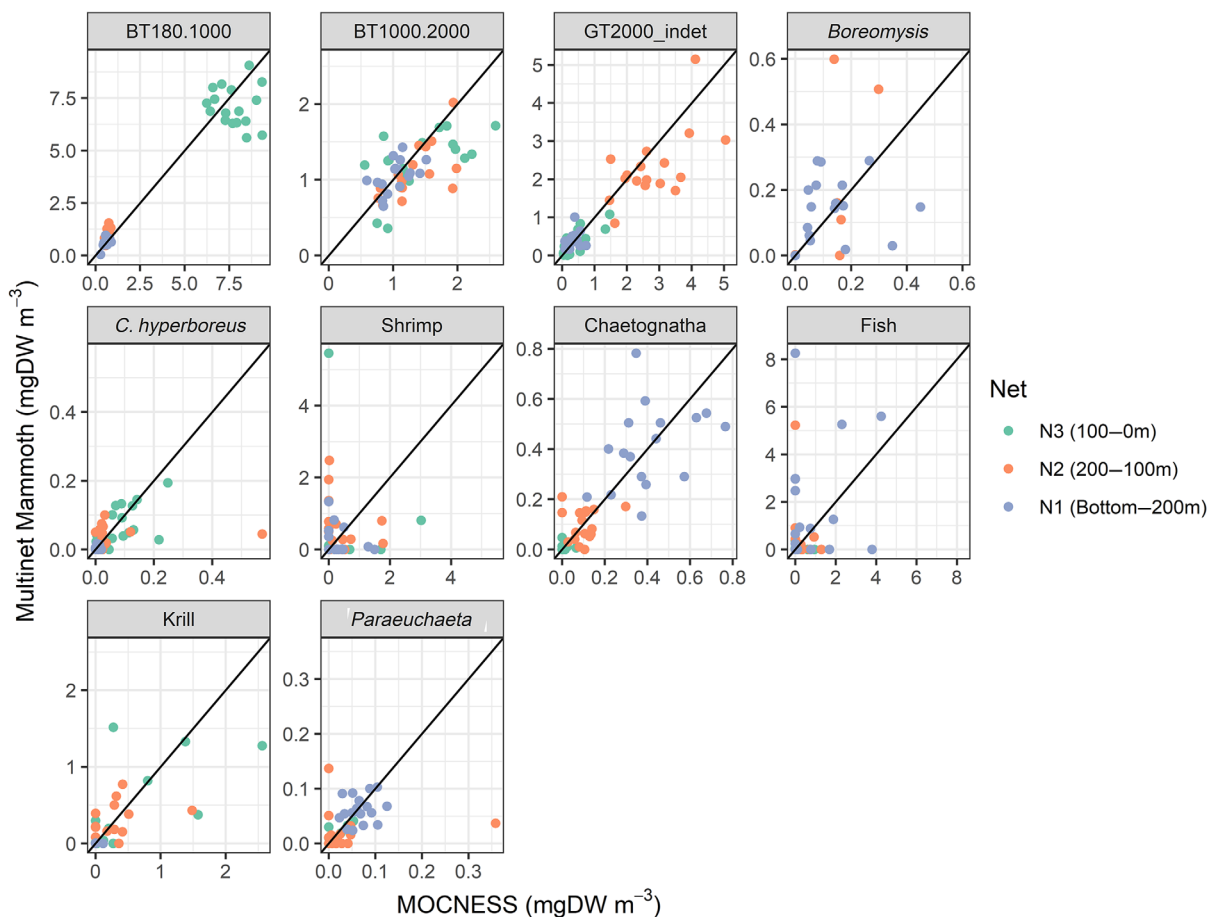
Given the relatively large biomass per individual fish compared to the smaller zooplankton, a large standard deviation is to be expected when few and a variable number of individuals are caught per deployment. The same reasoning also applies for the large standard deviation seen in biomass component Shrimps, consisting mainly of the genus *Sergestes* spp. (20 ind. in MOCNESS, in 15 Multinet) and *Pasiphaea* spp. (11 ind. in

MOCNESS, 17 in Multinet). With only 2 observations of Amphipods in the 34 integrated samples, this biomass component is not shown.

Due to the limited amount of water filtered by both equipment ( $1 \text{ m}^2$  opening) compared to gear more appropriate for measuring fish densities, 17 deployments appear, not surprisingly, to be too little to compare gear performance on species occurring with such low individual densities. For most other integrated biomass components, the mean was a close match and even with rather small standard deviations, it is not possible to say the equipment performed significantly different.

### Biomass of different components in different nets vs. equipment

Easing the level of data aggregation once more, we wanted to test if significant differences in gear sampling could be detected within the different depth strata sampled (Net 3: 0–100 m, Net 2: 100–200 m, Net 1: 200 to ~425 m). The chosen unit for this comparison was the calculated density ( $\text{mgDW m}^{-3}$ ) of each biomass component within each depth strata. The data show large variations in average biomass



**Fig. 3.** All data used in the statistical analysis visualized. Color of points show the different nets of the 17 pairs. The black line shows the 1 : 1 ratio of MOCNESS and Multinet biomass density. Note that axis range vary between plots.

**Table 4.**  $p$ -values of two-sided, pairwise  $t$ -tests with “BH” corrections for multiple testing (Benjamini and Hochberg 1995). All combinations of biomass components and net were tested against gear type. Significant results in bold ( $p_{\text{adjusted}} < 0.05$ ).

Biomass components	Net 3 (100–0 m)	Net 2 (200–100 m)	Net 1 (bottom to 200 m)
BT180.1000	0.571	<b>0.005</b>	0.972
BT1000.2000	0.704	0.129	0.972
GT2000_indet	0.704	0.341	0.773
<i>Boreomysis</i>	—	0.811	0.972
<i>Calanus hyperboreus</i>	0.773	0.811	0.811
Shrimp	0.972	0.811	0.811
Chaetognatha	0.972	0.972	0.972
Fish	0.811	0.811	0.704
Krill	0.811	0.972	0.811
<i>Paraeuchaeta</i>	0.972	0.811	0.811

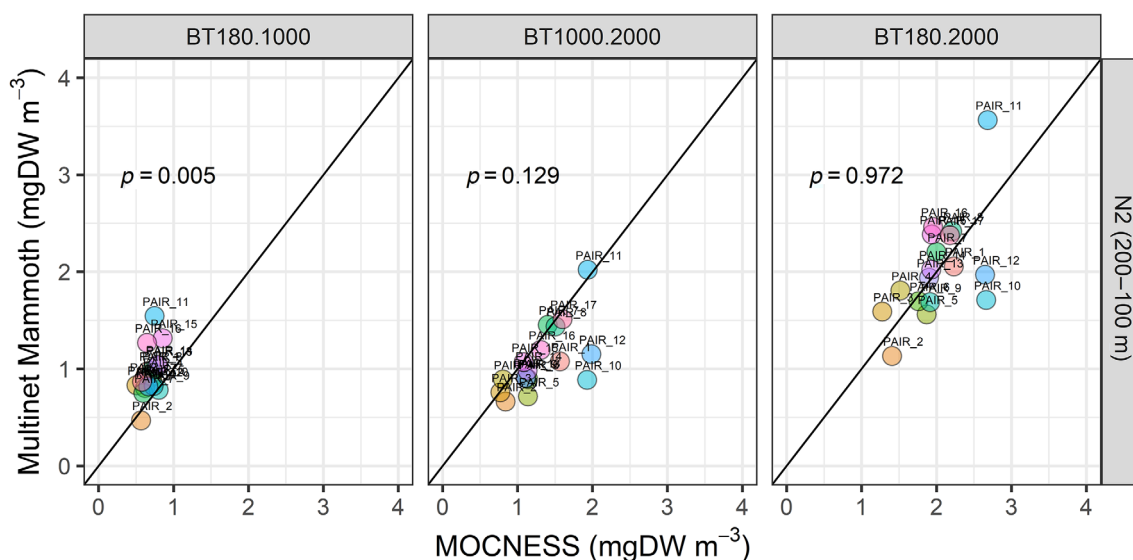
density between depth strata for most biomass categories (Fig. 3). The smallest size fraction was dominating the biomass in the upper net (100–0 m) while remaining biomass on the 2000- $\mu\text{m}$  screen (GT2000\_indet) after removal of other large components was dominating the middle net (200–100 m). In the lower net ( $\sim 425$ –200 m), mesopelagic fish and size fraction 1000–2000  $\mu\text{m}$  dominated, but the overall average density of dry weight was low. Almost all Chaetognaths were also found at this depth.

We found a significant variation between gears in the BT180.1000 size fraction for Net 2, while all other comparison were nonsignificant (Table 4). There is a risk of false positives

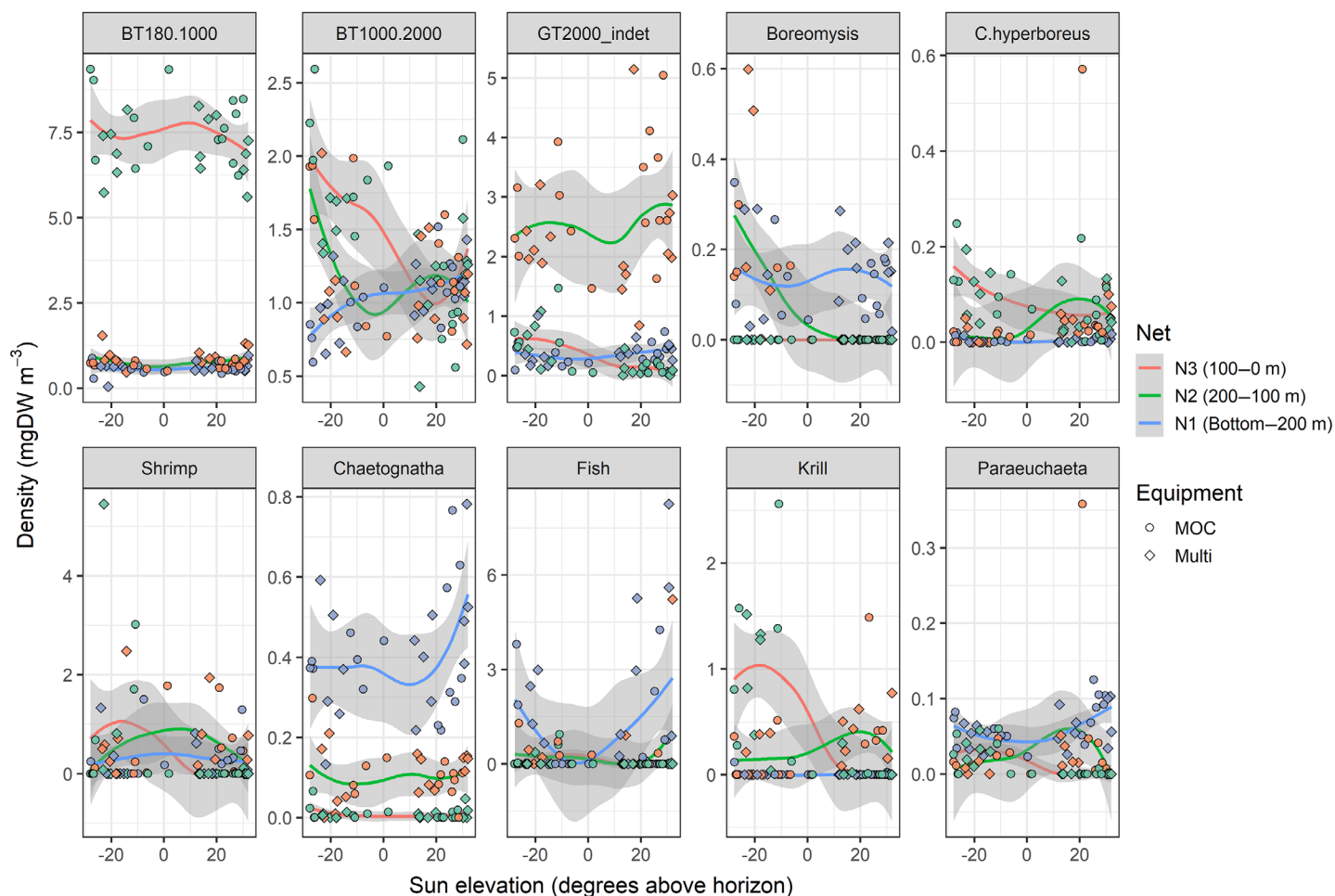
when performing as many as 30  $t$ -tests just by pure chance (10 biomass components  $\times$  3 nets), even with the Benjamini and Hochberg correction included for multiple testing. However, there are several things about this significant difference that makes it worthwhile to further investigate possible reasons: (1) Significant difference only appears in Net 2, not Net 1 and Net 3. (2) the mean biomass is significantly larger in the Multinet compared to the MOCNESS for the smallest size-fraction (BT180.1000), but the opposite, although not significant (Fig. 4a, b), is the case for the larger fraction (BT1000.2000). (3) If pooled (as before being size-fractionated), no significant relationship is apparent (Fig. 4c).

The first possible explanation for the observed difference could be related to the sampling gear itself. Both equipment have removable cod-ends that are numbered to match the corresponding net to ensure no mix-up once the cod-ends are removed from the net and transferred to the lab. This means that the same cod-end is used on the same net during every deployment. Both gears were thoroughly inspected, especially nets and cod-ends, for flaws before sampling started. If one or more small holes, smaller than 1000  $\mu\text{m}$ , were present in either the MOCNESS #2 cod-end and/or the #2 net attached to the MOCNESS frame, all samples would potentially lose small amounts mesozooplankton less than 1000  $\mu\text{m}$ . If this was the case, it might explain why only the #2 net had significant less BT180.1000 biomass in the MOCNESS compared to the Multinet. However, this would not explain that the MOCNESS appear to have more (although not significantly) of the BT1000.2000 fraction in the same net.

Another possible explanation is a human factor in the laboratory during the size fractionation of the remaining sample



**Fig. 4.** A closer look at biomass distribution in Net 2 plotted against each other. In the smallest size fraction, the Multinet catches significantly ( $p = 0.005$ ) more than the MOCNESS. For the middle fraction, the pattern is reversed, although not significantly ( $p = 0.129$ ). If one combines the two size fractions into BT180.2000, there is no significant differences in the paired  $t$ -test in Net 2 ( $p = 0.972$ ).



**Fig. 5.** Density measures of both equipment plotted against the sun elevation (degrees above the horizon) at time of sampling. Note that the y-axis varies depending on biomass component.

(here termed BT180.2000) into BT180.1000 and BT1000.2000 after all things larger than 2000  $\mu\text{m}$  were removed. Could the observed difference be due to different handling during fractionation? If more “force” or longer time is used while sieving the samples, one could expect more of the biomass to go through the 1000- $\mu\text{m}$  sieve, leaving a larger BT180.1000 and an accordingly smaller BT1000.2000 fraction. This could in theory explain the observed patterns. However, this seems unlikely since this result would only be apparent if the samples from one gear type and only from Net 2 would be treated differently. Also, since the sample processing on board was performed by two different teams (two persons each) working 6-h shifts around the clock, it is hard to see how one team performing the task differently could produce this error. Lastly, before sampling was started, both teams worked up a test-sample together to make sure they applied the same techniques and were following the same protocol for sample handling.

A last possible explanation worth exploring relates to how the opening/closing of nets are logged during stratified sampling.

The MOCNESS has a system where a digital response is sent to the surface when a net passes a mechanical lever as it opens/closes. Sometimes a small time-lag can be detected from when the signal to open a new net is sent to the response is received. Since the MOCNESS is moving continuously upwards ( $\sim 0.4 \text{ m s}^{-1}$ ) during sampling, this time-lag can also result in a small gap in the recorded depth profile of when a net is closed (signal sent) and the new one open (response received). Since the MOCNESS is constantly filtering water, even in the period between signal sent and response received, the depth of transition from one net to the other is assumed to be in the middle of the start and stop signal. The Multinet does not have such a response mechanism and the closing of one net and opening of the next is logged simultaneously when the signal is sent. Should there be a delay in the physical closing of a net after the signal is sent to the Multinet, this would result in the Multinet sampling a bit higher in the water column than what is logged. Since the density of BT180.1000 is much higher in the upper strata (Fig. 3), this could theoretically lead to a small bias towards sampling some of



the upper strata while recording it as part of the middle strata. However, for this to occur, the density of BT180.1000 in the few meters extra sampled above the strata (above 100 m for Net 2), would have to be significantly larger than in the depths below.

As for now, none of the above-mentioned reasons seem very likely apart from the possibility of there being one or more small holes in Net 2 that could explain the difference in BT180.1000.

### Day and night differences

Finally, we wanted to check if there were systematic differences between gears in sampling during day and night. The rationale behind this was that the two gear might affect individuals' gear avoidance differently by either visual cues or sound/vibration. Visual clues include own sensing of the gear (typically during day) or observation of bioluminescence (typically during night or in the deep). Sound and vibrations are likely independent of light, but individuals might react more to this when visual cues are scarcer due to darkness.

Pair 8 and 13 was removed from the data set since both the Multinet and the MOCNESS in the first case were assigned DAY (sun above the horizon) and to NIGHT (sun below the horizon) in the latter case.

For all combinations (10 biomass categories  $\times$  3 Nets  $\times$  [DAY + NIGHT] = 60 *t*-tests) none were significantly different, although Net 2 for the BT180.1000 fraction during DAY was borderline ( $p = 0.051$ ). That this category came out almost significant was to be expected based on the previous results (Fig. 4). However, care should be taken when interpreting these numbers since the number of available pairs available for analysis is rather low with nine pairs taken during DAY and only six during NIGHT. Even though there is a large variability for many biomass categories as a function of sun elevation (Fig. 5), nothing suggests that the two gear themselves sample differently, whether it is day or night.

### Discussion

Both the US-developed MOCNESS and the German-developed Multinet Mammoth have been widely used around the world for stratified zooplankton sampling. In order facilitate comparing samples taken by different institutions using either of the two equipment, as well as sound continuation of time series when a change of equipment is needed, comparison and intercalibration is important. The MOCNESS described here has previously been compared to the WP2 (Anonymous 1968) plankton net and found to sample more of the larger (GT2000) and less of the smaller (BT180.1000) zooplankton compared to the WP2 (Gjøsæter et al. 2000). For the middle size class, no differences were found.

In our case, the two multiple zooplankton sampling equipment MOCNESS and Multinet Mammoth, both with 1 m<sup>2</sup> opening and 180  $\mu$ m mesh sizes, were calibrated and pairwise compared using 3 vertically stratified nets during each of 17 deployments. Statistical analysis revealed no significant differences in terms of measured dry weight biomass for 10 different size-fractionated or species/taxa biomass categories in separate nets. The only exception was significantly more biomass for the 180–1000  $\mu$ m size-fraction in Net 2 of the Multinet Mammoth ( $0.946 \pm 0.2489 \mu\text{gDW m}^{-3}$ ) compared to the MOCNESS ( $0.689 \pm 0.096 \mu\text{gDW m}^{-3}$ ). However, when comparing both the BT180.1000 and the BT1000.2000 samples combined (as before being fractionated), this is no longer significantly different between gears. Our field experiment showed that the two gears sampled a wide range of plankton components similarly. Past, present, and future data acquisition with either equipment, should therefore be comparable for the analyzed species or groups assuming proper equipment calibration and under otherwise equal sampling conditions. Finally, we conclude that the continuation of the depth-stratified IMR time series on zooplankton will not be negatively impacted by the needed change of sampling equipment.

### Data availability statement

Data are stored at The Norwegian Marine Data Centre (NMDC).

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### Acknowledgment

The authors are grateful for the valuable help of the crew onboard R/VG.O. Sars during the cruise. This work was supported by Institute of Marine Research (Norway) and their monitoring programs by providing ship time and covering crew costs.

### Conflict of Interest

All authors declare that they have no conflicts of interest.

Submitted 15 March 2022

Revised 22 July 2022

Accepted 01 August 2022

Associate editor: Paul F. Kemp