Bachelor Thesis

Coastal and Marine Management

THE IMPORTANCE OF MESOZOOPLANKTON IN MANAGEMENT DECISIONS CONCERNING THE SOUTH-EASTERN WEDDELL SEA

ASSESSING THE CONTRIBUTION OF MESOZOOPLANKTON TO THE BIOMASS AVAILABILITY FOR HIGHER TROPHIC LEVELS AND ITS RELATION WITH SEA ICE



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Preface

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Abstract

The Southern Ocean is one of the harshest environments on the earth, yet it thrives with life. Krill is thought to be the pivotal organism in the Southern Ocean food web, as it channels the carbon produced by algae to higher trophic levels. However, mesozooplankton probably has a similar function in the Southern Ocean ecosystem. The ecosystem based management which is operated in the Southern Ocean focusses on krill and often neglects other carbon pathways. This study is set up to analyse how important mesozooplankton is in the food web of the south-eastern Weddell Sea, how its abundance and community structure is related to sea-ice and what this implies for the future management of the Southern Ocean. To answer these questions a combination of state-of-the-art research methods were used, including a size-class based approach in combination with C:N ratio analysis, bulk-stable isotope analysis, taxonomic analysis and literature studies. The biomass of mesozooplankton was found to exceed the biomass of krill in the Weddell Sea sampling area, and therefore has a higher contribution to the carrying capacity of the ecosystem. The most abundant mesozooplankton species was Calanoides acutus which corresponded with the size fraction with the highest biomass, namely 1000 μm. The undersea-ice mesozooplankton community (0-2 meter) was significantly different than in the 0-50 water depth layer. The C:N ratio was the highest in the 250 μm size fraction in the 0-2 meter. Regarding environmental variables, the interaction of chlorophyll a and sea-ice ridges best explained the species distribution of the 0-2 meter water layer. Species from the 0-2 meter depth stratum were found to be more dependent on sea-ice algae than the species in the 0-50 meter depth stratum. The health indicators of CCAMLR do not cover the whole ecosystem, as only high trophic level species are used. (Meso)zooplankton would be a good bottom-up health indicator, functioning as early warning indicator. Mesozooplankton and sea ice both fit in the WSMPA criteria and the general objectives of the WSMPA. It is concluded that mesozooplankton should be monitored and used as a health indicator for the ecosystem in the WSMPA.

Abbreviations

ANOSIM	Analysis of similarities
ANOVA	Analysis of variance
AWI	Alfred Wegener Institute
С	Carbon
CCAMLR	Commission for the Conservation of Antarctic Marine Living Resources
CEMP	CCAMLR Ecosystem Monitoring Program
ICEFLUX	Ice-ecosystem carbon flux in polar oceans
IUCN	International Union for Conservation of Nature
MSFD	Marine Strategy Framework Directive
MCA	Multi-criteria analysis
MPA	Marine Protected Area
Ν	Nitrogen
NMDS	Non metric multidimensional scaling
RMT	Rectangular Midwater Trawl
SCAR	Scientific Committee on Antarctic Research
SUIT	Surface and Under Ice Trawl
WMR	Wageningen Marine Research Institute
WSMPA	Weddell Sea Marine Protected Area

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1. Introduction

Antarctic Krill (*Euphausia superba*) is a major resource of the Southern Ocean and has a pivotal role in the Antarctic food web (Nilsson et al. 2016). Therefore *E. superba* is used as a criterion in most management decisions concerning this area. Yet, it is thought that mesozooplankton (250-4000 μ m) may be equally important in the food web of the Southern Ocean. This study is set up to analyse how important mesozooplankton is, in the food web of the south-eastern Weddell Sea and how its abundance and community structure is related to sea ice. The results will be used to make suggestions for future management decisions.

The Southern Ocean comprises the waters around Antarctica and covers approximately 10% of the global area covered by oceans (Constable et al. 2003). With approximately 2.8 million km², the

Weddell Sea is the largest marginal sea of Antarctica (Figure 1; (Encyclopaedia Britannica, 2008). Activities such as soil extraction, fishing and, in the last decades, tourism make the Antarctic an economically attractive area (Comba 2010). The economic value of the area is closely connected to its intrinsic value, as fishing and tourism are dependent on a healthy ecosystem (Chown et al. 2012). The fishery in the Southern Ocean mainly targets Patagonian toothfish (Dissostichus eleginoides), Antarctic toothfish (Dissostichus mawsoni), Mackerel icefish (Champsocephalus gunnari) and Antarctic krill (Euphausia superba). Increasing human activities and global warming endanger the ecosystems in the Southern Ocean. Climate change is altering the extent and thickness of sea ice



Figure 1: Map of the Southern Ocean indicating the position of the Weddell Sea (NASA, 2013)

in both the Arctic and the Antarctic seas rapidly (Garcia et al. 2016).

Sea ice is an important habitat for algae, mostly diatoms, which proliferate in and under the sea ice (Arndt & Swadling 2006; Arrigo & Thomas 2004). Growth of sea-ice algae precedes the phytoplankton bloom in spring and a second growth period of sea-ice algae is present in autumn, providing a source of food in periods when food in the water column is scarce (Arrigo & Thomas 2004). These sea-ice algae are eaten by zooplankton, such as copepods, amphipods and krill (Kiko et al. 2008; Froneman et al. 2000). Sea-ice algae have a prolonged growing season and are thought to be the main source of food for krill and other zooplankton during winter (Thomas & Dieckmann 2002). As it is predicted that sea ice will decline 24% in extent and 34% in volume at the end of this century (Arzel et al. 2006), possible changes in ecosystem functioning due to decrease in sea-ice are seen as a major future challenge in the management of the Southern Ocean (Constable et al. 2016; Nilsson et al. 2016).

Mesozooplankton such as copepods, pteropods and amphipods might be equally important as krill in the food web of the Weddell Sea (Flores et al. 2008; Van De Putte et al. 2006; Shreeve et al. 2005; Pakhomov et al. 2000; Boysen-Ennen et al. 1991), by forming a different carbon pathway than krill

(Figure 2; Murphy et al. 2013). Pakhomov et al. (2000) found that in the marginal ice zone of the Weddell Sea more than 50% of the biomass was derived from mesozooplankton. This suggests that a large proportion of the total carrying capacity is constituted by mesozooplankton, as at least half of the carbon transferred to higher trophic levels is channelled through mesozooplankton (Pakhomov et al. 2000). In this study carrying capacity is defined as the available biomass (mg C m⁻³) for higher trophic levels. Mesozooplankton provides an important food source especially for juvenile pelagic fish, myctophid fish, Antarctic silver fish (*Pleuragramma antarctica*), young Patagonian toothfish (*Dissostichus eleginoides*) and Antarctic toothfish (*Dissostichus mawsoni*) (Saunders et al. 2015; Pinkerton & Bradford-Grieve 2014; Pinkerton et al. 2013). Antarctic silver fish and myctophid fish species are crucial food sources for many different predators. As a multitude of trophic links exists between organisms (Figure 2), there is still uncertainty about the carbon pathway in the Antarctic food web (Nilsson et al. 2016; Pinkerton & Bradford-Grieve 2014).



Figure 2: An example of a Southern Ocean food web. The sizes of the squares, excluding detritus, show the proportion of the biomass of the groups to the power 0.1. The arrows indicate the direction of the organic carbon flow, excluding the flows back to detritus, for more clarity of the model (Pinkerton & Bradford-Grieve 2014).

The Commission for the Conservation of Antarctic Marine Living Resources (CCAMLR) has the responsibility for the conservation of the Antarctic marine ecosystems, including the sustainable harvest of marine resources (CCAMLR, 2016a). The CCAMLR convention entered into force in 1982, as a result of the Antarctic Treaty which was agreed on in 1959 (Secretariat of the Antarctic Treaty, 2013). The first objective of the Antarctic Treaty was peaceful coexistence in the Antarctic, followed by the objective for freedom of scientific investigation and the conservation of Antarctic flora and fauna (Secretariat of the Antarctic Treaty, 2013). Decisions within the CCAMLR are taken by the CCAMLR commission based on recommendations given by the scientific committee. Both consist of members from all 24 states and the European Union, which have signed the CCAMLR convention (CCAMLR, 2016d). A special characteristic of the CCAMLR convention system is that the commission has the obligation to take the advice from the scientific committee into account (CCAMLR, 2016e).

In the 1990's, CCAMLR adopted the ecosystem based approach to manage the living resources in the Southern Ocean (Constable 2011; Nilsson et al. 2016). In accordance with this approach the CCAMLR Ecosystem Monitoring Program (CEMP) was set up, to detect changes in the ecosystem caused by harvesting marine living resources in the convention area (CCAMLR, 2013a). CEMPs health indicators are especially krill dependent species. Krill has a pivotal role in the Southern ecosystem as it is prey for many birds, penguins and whales (Nilsson et al. 2016). However, by taking only krill into account, CCAMLR focusses on a single trophic pathway and neglects the contribution of other trophic groups to the carbon availability for higher trophic levels (Constable et al. 2016). As ecosystems are complex, it can be questioned how useful a single trophic pathway is to deduce ecosystem functioning (Constable et al. 2016; Nilsson et al. 2016). Krill abundance is highly variable between years, and a 10-fold variation in krill abundance can be present (Shreeve et al. 2005). In years where krill is less abundant, mesozooplankton achieves higher biomass and becomes a major component in the diet of higher tropic levels (Shreeve et al. 2005). Additionally, future changes in sea-ice extent and volume through climate change are not taken into account by CCAMLR. Although the importance of sea ice has been established for krill, for other zooplankton it remains largely unclear whether they are dependent on sea ice (Jia et al. 2016; Wallis et al. 2016; Kramer et al. 2011; Krapp et al. 2008).

It is expected that the south- eastern Weddell Sea will play a crucial role for sea-ice dependent organisms in the future (Teschke et al. 2013), as convergent hydrological and meteorological conditions cause the sea ice to drift into the Weddell Sea (Brierley & Thomas 2002). Additionally, the south-eastern part of the Weddell Sea has a high biodiversity which is even comparable to tropical ecosystems in some areas (Brey et al. 1994). In terms of commercial fishery activities, the south-eastern part of the Weddell Sea remains relatively unexploited as only a minor toothfish fishery is established in this sector (Teschke et al. 2016b).

Germany took the lead in an initiative to realise an MPA in this area of the Weddell Sea, the WSMPA. The WSMPA fits in the CCAMLR conservation Measure 91-04 (2011), which was developed to set up a system of MPAs in the Southern Ocean, with the aim to conserve marine biodiversity (Teschke et al. 2016a). This MPAs support the main objective of the CCAMLR convention, namely the "the conservation of Antarctic marine living resources, including their rational use " (Teschke et al. 2016a). The WSMPA will become a no take zone and a reference area for scientific research (European Commision 2015). To delineate the exact WSMPA area several abiotic and biotic criteria are used. The Antarctic krill (*Euphausia superba*), as a commercial important species and a crucial link in the carbon transfer to higher trophic levels, was used as one of the major criteria (Teschke et al. 2016b). However, in order to serve as a reference area and to be able to discriminate between natural and anthropogenic effects, quantification of the amount of carbon in different pathways is crucial, as it allows for the detection of shifts in the relative importance of each pathway (Constable et al. 2016).

The Alfred-Wegener-Institute (AWI) in Germany and the Wageningen Marine Research Institute (WMR) in the Netherlands aim to get a better understanding about the Southern Ocean ecosystems. The Ice-ecosystem carbon flux in polar oceans (Iceflux) project group of the Alfred-Wegener-Institute was specifically set up to investigate the importance of sea ice for the pelagic food web of the Polar Oceans. The aim is to gain understanding on how the food web will respond to the declining sea ice. An important goal is to fill gaps of currently missing knowledge on food web structure and the role of sea ice towards sustainable approaches of fisheries management and nature conservation in the Southern Ocean (Alfred-Wegener-Institut, 2016).

As mesozooplankton possibly fulfils a major function in the Weddell Sea ecosystem, as source of carbon for higher trophic levels, this has implications for the management of the Southern Ocean, especially the Weddell Sea. Since management is mainly based on krill and ignores alternative carbon pathways, it can be questioned how "ecosystem based" the management approach actually is. In addition, possible declines in sea-ice extent, which may cause shifts in community structure as a result of decreased food availability or available habitat (David et al. 2017; Flores et al. 2014), are not taken into account in the current management framework of CCAMLR or in the delineation of the WSMPA. Knowledge about the food web structure in combination with the amount of biomass available in different pathways of the food chain will provide valuable clues about the functioning of the ecosystem and could serve as baseline for the detection of shifts in ecosystem functioning. In addition, further elaboration of dependency of organisms on sea ice is essential for predicting effects of expected sea-ice decline on the Antarctic ecosystem and its carrying capacity (David et al. 2017; Schaafsma et al. 2016; Flores et al. 2014; Flores et al. 2011).

1.2. Research Goal

The goal of this research is to:

- i) identify the contribution of mesozooplankton to the carrying capacity for higher trophic levels in the food web of the south-eastern Weddell Sea and compare it to krill, by using a combination of a size-based, C:N ratio -based approach.
- ii) identify the influence of sea ice on mesozooplankton abundance, biomass, size based community structure and taxonomic community structure, based on bulk stable isotope analysis and the environmental parameters sea-ice coverage, sea-ice thickness, number of sea-ice ridges, vertical proximity to sea ice, temperature, salinity and chlorophyll *a* concentration.
- iii) to suggest improvements to the current management approach adopted by CCAMLR and to give recommendations for the criteria used to establish the WSMPA.

This study aims to contribute towards the improvement of ecosystem based management of the Southern Ocean, by better understanding and quantifying an overlooked component of the Antarctic food web.

1.3. Research Question

Main question:

How important is mesozooplankton in the food web of the south-eastern Weddell Sea, including its relation to sea ice, and what do the findings of this study implicate for future management decisions of CCAMLR and the WSMPA project team?

Sub-questions:

- 1. What is the contribution of mesozooplankton to the carrying capacity of the south-eastern Weddell Sea in comparison with krill?
- 2. Can the size class and taxonomic mesozooplankton community structure and spatial distribution be related to sea ice?
- 3. Should mesozooplankton and sea ice be used as criteria for management decisions by CCAMLR and the WSMPA project team?

This introduction is followed by the materials and methods section. It includes a short sample area description, a part about the sample analysis and the statistical analysis section. After the materials and methods chapter, the results are itemised in ecological and management findings. The discussion is also split up in ecology and management and the conclusion section follows-up. The report is closed with recommendations for further research. After the references in the appendices, information about the sample stations can be found.

2. Materials & Methods

2.1. Area description

The Southern Ocean is separated from the other oceans by the Polar Front (Constable et al. 2003). It is a very variable environment, with extensive seasonal changes. It cannot be seen as one ecosystem as there are huge regional differences depending on various, dominant abiotic factors (Constable et al. 2003). The Antarctic area has 14 marginal seas. The Weddell Sea sector (60°W to 20°E) is the largest of these marginal seas (Encyclopaedia Britannica, 2008). Its borders are defined by the Ronne-Filchner ice shelf in the south and the Atlantic-Indian Ridge in the north. The maximal depth of the Weddell Sea is 5000 meter (Teschke et al. 2013). An important feature of the Weddell Sea is the Weddell Gyre rotating clockwise within the marginal sea. It plays a major role in global thermohaline circulation (Teschke et al. 2013). One of the most distinct features of the Weddell Sea is the large extent of sea-ice cover and the high seasonal changes of the sea-ice cover. It is a highly productive area and at the same time one of the most variable regions of primary production in the Southern Ocean (Constable et al. 2003). The south-eastern Weddell Sea has especially high biodiversity in comparison with other polar regions (Brey et al. 1994).

2.2. Data collection

Samples were collected in the south-eastern Weddell Sea during research cruise PS89 on board of RV *Polarstern*, between 12 December 2014 and 20 January 2015 (Figure 3; Appendix 1; Flores et al. 2015).



The zooplankton samples were taken in the 0-2 meter depth stratum with a Surface and Under-Ice Trawl (SUIT) and in the 0-50 meter depth stratum with the Rectangular Midwater Trawl (RMT). The SUIT was used to sample the upper two meters of the water column under the sea ice as well as in open water. As the SUIT sampled at the side instead of in the wake of the ship, it was able to collect the zooplankton in a relative undisturbed environment. The RMT sampled the pelagic water column from 0-50 meter. Both Trawls had sensors which measured the volume of water sampled. A sensor array mounted in the SUIT measured environmental parameters, such as temperature, salinity, chlorophyll *a* concentration and depth. The sea-ice cover, sea-ice ridges and sea- ice thickness values were derived from Müller (2016), who was calculating this data for her Bachelor thesis. For further details about the sampling procedure, see Flores et al. (2015)

Zooplankton catches from both the SUIT and RMT were split into two equal parts using a plankton splitter. One half was preserved in a 4% hexamine-buffered formaldehyde-sea water solution for analysis of taxonomic composition. The other half was wet sieved with water over stacked sieves (125, 250, 500, 1000, 2000, and 4000 μ m mesh size) to split the samples into size fractions (Veit-köhler et al. 2013). These fractions were then oven dried in petri-dishes and stored at -80°C for further use.

The mesozooplankton samples of the SUIT plankton net (0.3mm mesh size) and RMT 1 net (0.33 mm mesh size) were analysed in the laboratory, at the AWI, via dry weight measurements, carbon and nitrogen analysis, bulk stable isotope analysis and taxonomic- and size-based classification. Krill data from the SUIT krill net (7mm mesh size) and RMT 8 net (4.5mm mesh size), from the same cruise as the mesozooplankton samples, have already been analysed in terms of size and taxonomy. The data were used to compare the available carbon of Antarctic krill and mesozooplankton, in order to establish the contribution of each group to the carrying capacity.

2.3. Sample analysis

Taxonomy

The species composition and abundances of mesozooplankton directly underneath the sea ice (0-2 meter depth stratum) and in the 0-50 meter depth stratum were assessed. Samples were fractionated with a Motodo plankton splitter (1/4 - 1/64 of the original sample size) and a subsample of the original sample was analysed for species composition and abundance. Species were identified to the lowest taxonomic level possible and abundances were scaled up afterwards to the original sample size by multiplying the abundances by the subsampling fraction. Data was standardised by dividing the abundances with the water volume filtered during the haul. Station 79-1 (Appendix 1) contained a large amount of *Pheocystis* and the amount of fibres present in the sample made it unworkable. Therefore, this station was not assessed for species composition and abundance.

Size-class based approach

For the mesozooplankton biomass, the C:N ratio analysis and the bulk stable isotope analysis, a sizeclass based approach was chosen. Zooplankton grows several orders of magnitude in their lifetime, thereby passing several trophic levels. As the larger organisms feed on the smaller ones it can be helpful to use a size-class-based approach to determine the role of mesozooplankton in the food web (Tarling et al. 2012). In addition, is the size spectra and changes in the size spectra of a zooplankton community informative about the health of the marine ecosystem (Gorokhova et al. 2016; Connors et al. 1978). The four size classes 250-500 $\mu m;$ 500-1000 $\mu m,$ 1000-2000 $\mu m,$ 2000-4000 μm mesh size were used.

Biomass

The biomass is defined as carbon per cubic meter. For mesozooplankton, each size fraction was freeze dried for 48 hours, weighed to the nearest microgram and homogenised. From the homogenised material three replicates of 0.5-1.5 mg were analysed for carbon and nitrogen with a Carlo Erba CN analyser (HEKAtech GmbH, Germany). Carbon biomass was calculated for each size fraction separately by first calculating the mean proportion of Carbon from the three replicates. Then the dry weight of the corresponding size class was multiplied by the mean proportion of Carbon to obtain the biomass of the size class (mg C). The amount of Carbon within each size class was then divided by the volume of sea water filtered during sampling to obtain the Biomass per m³ (mg C m⁻³). The biomass of the 250, 500, 1000 and 2000 μ m size fractions were summed to obtain the mesozooplankton biomass per station.

Length to dry-weight relationships from Mizdalski (1988) were used to calculate the dry weight of the krill catch. Carbon and Nitrogen analysis data of krill from the PS89 cruise were used to convert the dry weight into biomass (mg C). The biomass was divided by the volume of sea water, filtered through the haul per station, to obtain the biomass per m³ (mg C m⁻³).

C:N ratios

The food quality of each size fraction was determined by analysing the C:N ratio. Proteins have a C:N ratio around 3, which increases with increasing lipid content (Harris et al. 1986). Therefore C:N ratio is often used as an indicator of the protein/lipid ratio in the body (Donnelly et al. 1994). As lipids have a higher energetic value than proteins (Clarke 1980), lipid rich food is considered as high quality food. Furthermore, as high lipid content is an indicator of a fat reserve, used to overcome periods of food scarcity, high lipid content suggests that the organism is in good condition (Harris et al. 1986).

As explained for the mesozooplankton biomass three homogenised replicates of 0.5-1.5 mg were analysed for Carbon and Nitrogen with a Carlo Erba CN analyser (HEKAtech GmbH, Germany). The C:N ratios were calculated for each replicate by dividing the carbon value with the nitrogen value.

Bulk stable isotope analysis

From the bulk stable isotope analysis the δ^{15} N value and the δ^{13} C value were retrieved. The δ^{15} N was used to indicate trophic level differences between the size fractions, as δ^{15} N accumulates in higher trophic levels relative to their prey (Tarling et al. 2012). The δ^{13} C value was used to analyse the dependency of mesozooplankton on ice algae. In the sea-ice environment, carbon availability is often limited and then results in a higher proportion of the heavy ¹³C isotope over the lighter ¹²C isotope (Kohlbach et al. 2016). Thus heterotrophic production from within the sea ice (ice-algae) has a higher δ^{13} C than heterotrophic pelagic production (phytoplankton) (Kohlbach et al. 2016).

From each size fraction 3 replicates were analysed with a continuous flow isotope ratio mass spectrometer Delta V Plus, interfaced with an elemental analyser (Flash EA 200 series) and connected to a Conflo IV interface (Thermo Scientific Corporation, Germany). The isotopic ratios

were expressed as parts per thousand (‰) in the δ notation, according to the following equation from Coplen (2011):

$$\delta_{x} = [(R_{sample}/R_{Standard})-1] \times 1000$$
 (2)

Where x represents the heavy carbon isotope δ^{13} C or the heavy nitrogen isotope δ^{15} N. R_{sample} is the δ^{13} C or δ^{15} N in the sample relative to R_{standard}. The international Vienna Pee Dee Belemnite standard (C) and atmospheric N₂ were used as reference (R_{Standard}) (Kohlbach et al. 2016; Veit-köhler et al. 2013). Calibration of the stable isotope measurements was done according to the protocol by Brand et al. (2014) with the reference materials USCG40 and USCG41a.

2.4. Statistical analysis

The statistical analysis of this study was done with the statistical programming software R (The R Foundation, 2016).

Krill biomass vs mesozooplankton biomass

The relative biomass (%) was normally distributed in the 0-2 meter depth stratum (Shapiro-Wilkinson normality test, d f= 10, p= 0.057), but not in the 0-50 meter depth stratum (Shapiro-Wilkinson normality test, df = 16, p = 0.016). Therefore, the differences in relative biomass (in %) of krill and mesozooplankton was assessed with an independent sample t-test for the 0-2 m depth stratum and a Mann-Whitney U-test for the 0-50 meter depth stratum. Variances were equally distributed for both depth strata (Levene's test for equality of Variances, F = 0.000, p = 1.000). Station 41-1 was excluded from the analysis, as krill data was missing for this station.

Differences in the total biomass in the 0-2 meter water column and the 0-50 meter water depth stratum column were assessed with a Mann-Whitney U-test, as the data was not normally distributed (Shapiro-Wilkinson normality test, df=14, p = 0.009). To discover if total krill and mesozooplankton biomass differed between the depth strata, two Mann-Whitney U-tests were performed, because krill biomass was not normally distributed (Shapiro-Wilkinson normality test, df = 14, p < 0.001) and mesozooplankton biomass was also not normally distributed (Shapiro-Wilkinson normality test, df = 14, p = 0.010). Station 41-1 (Appendix 1) was excluded from the analysis, as krill data was missing for this station.

Size fractions per depth stratum

To identify which size fractions had the highest contribution to mezozooplankton biomass within each depth stratum, the relative biomasses of the size fractions 250, 500, 1000, and 2000 μ m were compared with a Tukey HSD test. Although the ANOVA assumptions were violated, a Tukey HSD test was used, as it works independent from the ANOVA and does not require normality or equal variances.

C:N ratio mesozooplankton

The C:N ratio data for both depth strata violated the assumptions for an ANOVA. The C:N ratio was not normally distributed in the 0-2 meter depth stratum (Kolmogorov-Smirnoff normality test, df = 58, p <0.001) and in the 0-50 meter water column (Kolmogorov-Smirnoff normality test, df = 91, p <0.001). The variances were equal between the size fractions in the 0-2 meter depth stratum (Levene's test for Homogeneity of Variances, F= 0.876, p = 0.459), but were unequal between the

size fraction in the 0- 50 meter depth stratum (Levene's test for Homogeneity of Variances, F= 7.411, p < 0.001). Therefore, a Tukey HSDs test was performed, to assess differences in the C:N ratio between the size classes 250, 500, 1000 and 2000 μ m, for both depth strata.

Zooplankton community

Analysis of similarity (ANOSIM) was conducted on a Bray- Curtis dissimilarity matrix of the log(x+1) transformed community data with the 0-2 and 0-50 meter depth strata as grouping factor, to discover whether the zooplankton communities differ between depth strata. For the 0-50 meter stratum, an additional ANOSIM was performed with presence or absence of ice as grouping factor to discover if the presence of sea ice has an influence on the zooplankton species present in these depth strata. A NMDS plot was constructed to visualize the results of the ANOSIM analysis. In addition, a Cluster-analysis was performed on the Bray-Curtis dissimilarity matrix between the stations to be able to determine which stations are more similar in community structure. Ward linkage method was used as it performs best in many situations (Ferreira & Hitchcock 2009).

Environmental parameters

Bioenv analysis was performed on the species abundance data of the 0-2 meter depth stratum and the environmental parameters measured while trawling: chlorophyll a, sea-ice thickness, sea-ice cover, number of sea-ice ridges, salinity and temperature. All environmental variables were normalised to obtain a consistent scale. Normalisation was achieved by subtracting the mean of each parameter from a single observation of that parameter and dividing by the standard deviation. The Bioenv was run using a Bray-Curtis dissimilarity matrix and spearman rank correlation to determine which environmental parameters best explain the species distribution. Bioenv analysis estimates the subset of environmental parameters that have the highest correlation with the abundance data, thus indicative of the environmental parameters that best explain the distribution of species (Clarke & Ainsworth 1993). A mantel test was used to test the association of the rank correlation of the species abundance, with the selected subsets of environmental parameters. The p value was calculated using 999 iterations.

Bulk stable isotope

To identify differences in δ^{15} N isotopic values for the size fractions 250, 500, 1000, 2000 and 4000 µm for the 0-50 meter depth stratum and the size fractions 500, 1000, 2000, 4000 µm for the 0-2 meter depth stratum a Tukey HSD test was performed for each stratum, as the assumptions for a parametric test were violated. For the 0-50 meter water column the δ^{15} N isotopic value were not normally distributed (Shapiro-Wilkinson normaility test, df= 43, p = 0.004). In the 0-50 meter depth stratum the variances for δ^{15} N were equal among groups (Levene's test of homogeneity of variances, F= 1,980, p = 0.117). In the 0-2 meter depth stratum the δ^{15} N values were not normality test, df = 25, p = 0.007) and the variances were equal among groups (Levene's test for homogeneity of variances, F = 0.565, p = 0.691). The 250 µm size fraction was excluded in the 0-2 meter depth stratum, as the δ^{15} N value was not reliable due to insufficient sample materials in the BSI analysis.

To identify differences in δ^{13} C isotopic values for the size fractions 250, 500, 1000, 2000 and 4000 µm for the 0-50 meter depth stratum and the size fractions 500, 1000, 2000, 4000 µm for the 0-2 meter depth stratum an ANOVA was performed for each stratum. In the 0-2 meter water column the δ^{13} C

isotopic values were normally distributed (Shapiro-Wilkinson normaility test, df= 25, p = 0.092) and the variances were equal among groups (Levene's test for homogeneity of variances, F= 1.923, p = 0.126). For the 0-50 meter water column the δ^{13} C isotopic values were normally ditributed (Shapiro-Wilkinson normality test, df= 43, p = 0.156) and the variances were equal among groups (Levene's test for homogeneity of variances, F = , 0.768 p =0.558). The 250 μ m size fraction was excluded in the 0-2 meter depth stratum, as the δ^{13} N value was not reliable due to insufficient sample materials in the BSI analysis.

Independent sample t -test were performed to identify differences in the δ^{13} C isotopic values per size fraction, between the 0-2 meter and 0-50 meter depth strata.

The δ^{13} C isotopic values in the 500 µm size fraction were normally distributed (Shapiro- Wilkinson normality test, df = 14, p = 0.163) and the variances were equal among groups (Levene's test for homogeneity of variances, F = 1.903, p = 0.193).

The δ^{13} C isotopic values in the 1000 µm size fraction were normally distributed (Shapiro- Wilkinson normality test, df = 14, p = 0.140) and the variances were equal among groups (Levene's test for homogeneity of variances, F = 0.140, p = 0.714).

The δ^{13} C isotopic values in the 2000 μ m size fraction were normally distributed (Shapiro- Wilkinson normality test, df = 13, p = 0.459) and the variances were equal among groups (Levene's test for homogeneity of variances, F = 0.049, p = 0.828).

The δ^{13} C isotopic values in the 4000 µm size fraction were normally distributed (Shapiro- Wilkinson normality test, df = 14, p = 0.376) and the variances were equal among groups (Levene's test for homogeneity of variances, F = 0.988, p = 0.340).

2.5. Management analysis

The question if mesozooplankton and sea ice should be incorporated in management decisions over the Southern Ocean was mainly based on the ecological findings of this study. In addition, in order to be able to suggest if and how mesozooplankton and their possible relation to sea ice should be incorporated in the management of the Weddell Sea, the current management situation of CCAMLR and the WSMPA project team was analysed via literature study first.

CCAMLR

It was summarized how CCAMLR is currently covering krill and mesozooplankton in their research from the scientific committee and in the CEMP health indicators. This was done by looking at the tasks from the research working groups of CCAMLR, at the conservation measures and at the CEMP health indicator species, including their diets. Via the criteria suggested by Hilty & Merenlender (2000) the suitability of mesozooplankton as a health indicator and the suitability as adequate complementation for the CCAMLR ecosystem management was analysed. Hilty & Merenlender (2000) used 9 published articles from which they choose criteria for health indicator species which were stated in more than one reference and these criteria are still widely used (Hemraj et al. 2017; Alessandro et al. 2016; Stokes et al. 2016). Furthermore it was analysed how the current CCAMLR framework is handling future sea ice declines at the moment.

<u>WSMPA</u>

The history of the MPAs in the CCAMLR convention area was summarized, as well as the method developed for the delineation of the best MPA areas. The general criteria which an MPA should include were stated to see if mesozooplankton or sea ice does fulfil them. Also the specific criteria used for the delineation of the WSMPA were summarized to determine if and how zooplankton and sea ice are used as criteria in the establishment of the WSMPA planning area at the moment. The general objectives for the future of the WSMPA are stated to determine if mesozooplankton and sea ice would fit into these objectives.

3. Results

3.1. Ecology

The relative biomass of mesozooplankton (mean = $82.47\% \pm 15.50\%$, n=8) was significantly higher in the 0-50 meter depth stratum in comparison to the krill biomass (mean = $17.53\% \pm 15.50\%$, n=8) (Mann-Whitney U -test, U =64, p <0.001; Figure 4). In the 0-2 meter water column there was no significant difference between the relative biomass of mesozooplankton (mean = $42.08\% \pm 37.07\%$, n=5) and krill (mean = $57.92\% \pm 37.07\%$, n=5) (Independent sample t-test, df = 8, t = 0.675, p= 0.519; Figure 4).



Figure 4: Relative biomass of mesozooplankton in comparison with krill, in the 0-2 meter and 0-50 meter depth strata. The error bars represent the 95% confidence interval.

Total krill biomass did not significantly differ between the 0-50 depth stratum (range 0.02-1.22 mg m⁻³) and 0-2 depth stratum (range 0.02-0.40 mg m⁻³) (Mann-Whitney U-test, U = 15, p = 0.524). The total biomass of mesozooplankton was significantly higher in 0-50 depth stratum (range 0.10-5.18 mg m⁻³) than in the 0-2 meter depth stratum (range 0.02-1.77 mg m⁻³)(range (Mann-Whitney U-test, U = 4, p = 0.019). The combined total biomass of krill and mesozooplankton was significantly higher in the 0-50 meter depth stratum in comparison with the 0-2 meter depth stratum (Mann- Whitney U-test, U = 5, p = 0.041). The combined total biomass at all 0-2 meter depth stratum stations covered by sea ice was very low, with a minimum of 0.06 mg m⁻³ at station 29-1 and a maximum of 0.47 mg m⁻³ (Figure 6). The highest biomass encountered in the 0-2 meter depth stratum was at station 27-6, with 1.90 mg m⁻³ (Figure 6). At this station, no sea ice was present. The sea-ice zone stations in the 0-50 meter depth stratum had a maximum biomass of 5.26 mg m⁻³ at station 27-5 and a minimum biomass of 1.21 mg m⁻³ at station 66-2 (Figure 5). The lowest biomass in the 0-50 meter depth stratum was at station 79-1, with 0.13 mg m⁻³ (Figure 5).

Seven of the eight 0-50 meter stations (RMT) had a higher relative mesozooplankton biomass in comparison with krill biomass and one was 50 percent (Figure 5).



Figure 5: Overview map of the 0-50 meter stations and the relative biomass of mesozooplankton in comparison with krill

Two of the five 0-2 meter stations (SUIT) had a higher relative mesozooplankton biomass in comparison with krill and three had a higher relative krill biomass (Figure 6).



Figure 6: Overview map of the 0-2 meter stations and the relative biomass of mesozooplankton in comparison with krill

The relative mesozooplankton biomass of the 1000 μ m size fraction was significantly higher than the 250, 500 and 2000 μ m size fractions, in the 0-50 meter depth stratum (Tukey HSD, p < 0.001 between 1000 μ m and all other size fractions; Figure 7). There were no significant differences in relative biomass between the size fractions of the 0-2 meter depth stratum (Figure 7). The mean biomass per depth strata is given in Table 1.



Table 1: Mean biomass per size fraction

	Size fraction (µm)	Mean total biomass of the 0-50 meter (mg m ⁻³)	Mean total biomass of the 0-2 meter (mg m ⁻³)
	250	0.03 ± 0.04	0.01 ± 0.02
	500	0.19 ± 0.23	0.03 ± 0.05
otn	1000	1.48 ± 1.35	0.32 ± 0.70
0-50 meter	2000	0.26 ± 0.33	0.02 ± 0.01
0-2 meter			

Figure 7: Relative contribution of the four mesozooplankton size classes to the total biomass. The different letters indicate significant differences, whereas the same letter indicates no significant difference between the size fractions. The error bars represent the 95% confidence interval.

The mean mesozooplankton C:N ratio is higher in the 0-2 meter depth stratum (4.88 ± 1.35) in comparison with the 0-50 meter depth stratum was (4.32 ± 0.94). In the 0-50 meter depth stratum, the C:N ratio of the 1000 μ m size fraction was significantly higher than the 500 μ m size fraction (Tukey HSD, p = 0.005) and the 2000 μ m size fraction (Tukey HSD, p = 0.001; Figure 8). In the 0-2 meter depth stratum, the 250 μ m size had a significantly higher C:N ratio than the 500 μ m size fraction (Tukey HSD, p < 0.001), the 1000 μ m size fraction (Tukey HSD, p < 0.001) and the 2000 μ m size fraction (Tukey HSD, p < 0.001) and the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p < 0.001). In addition, the 500 μ m size fraction was significantly higher than the 2000 μ m size fraction (Tukey HSD, p = 0.025). It was not tested for differences between the 0-2 meter depth stratum and the 0-50 meter depth stratum.



The subclass copepod dominated the total abundance of mesozooplankton species with 82%. In the total species abundance of the 0-50 meter depth stratum 81% were copepods and in the 0-2 meter depth stratum 94% were copepods. Calanoides acutus dominated the mesozooplankton, followed by Metridia sp. and Ctenocalanus sp. (Table 2).

All stations			0-50 meter (RMT)				0-2 meter (SUIT)						
(n= 13)		all (n=8)		with sea ice (n=4)		without sea ice		all (n=5)		with sea ice (n=4)		without sea ice	
						(n=4)						(n=1)	
Calanoides	39%	Calanoides	39%	Calanoides	66%	Calanoides	32%	Calanoides	41%	Paralabidocera	80%	Calanoides	49%
acutus		acutus		acutus		acutus		acutus		antarcica		acutus	
<i>Metridia</i> sp.	12%	<i>Metridia</i> sp.	12%	Metridia sp.	9%	Metridia sp.	13%	Ctenocalanus	18%	Calanoides	7%	Ctenocalanus	22%
								sp.		acutus		sp.	
Ctenocalanus	9%	Ctenocalanus	9%	Ctenocalanus	4%	Ctenocalanus	10%	Paralabidocera	18%	Stephos	3%	Copepodites	10%
sp.		sp.		sp.		sp.		antarctica		longipes			
Rhincalanus	6%	Rhincalanus	6%	Calanus	3%	Rhincalanus	7%	Copepodites	9%	Copepodites	2%	Oithona sp.	9%
gigas		gigas		propinquus		gigas							
<i>Oncea</i> sp.	6%	Oncea sp.	6%	Paraeuchaeta	2%	Oncea sp.	7%	Oithona sp.	7%	Limacina	2%	Stephos	2%
				sp.						helicina		longipes	

Table 2: Showing the most abundant species/families of mesozooplankton in descending order.

Overall, *Calanoides acutus* dominated in the 0-50 meter depth stratum and was mainly found in 1000 μm size fraction. In the 0-2 meter depth stratum with sea ice *Paralabidocera antartica* dominated which was mainly found in the 500 μm size fraction.

The zooplankton community differed significantly between the 0-2 meter depth stratum and the 0-50 meter depth stratum (ANOSIM, R = .699, p = 0.001). The variance between the 0-2 meter and 0-50 depth strata was higher than the variance within the 0-2 meter and 0-50 meter depth strata. The NMDS plot visualizes the outcome of the ANOSIM analysis (Figure 9). *P. antarctica, S. longipes* and Euphausids were associated with the 0-2 meter depth stratum. *C. acutus, C.popinquus, Metridia sp., Paraechaeta sp.* were associated with the 0-50 meter depth stratum.

All stations from the 0-2 meter depth stratum that are clustered together in the NMDS plot were stations where sea ice was present. The 0-2 meter station (27-6), were sea ice was absent, is located near the 0-50 meter stations in the NMDS plot (Figure 9).



Figure 9: Non-metric multidimensional scaling plot showing the different community structures of the SUIT and the RMT.

The same result was revealed via a cluster analysis (Figure 10). The right cluster was comprised of the 0-2 meter depth stratum stations where sea ice was present. The left cluster consisted of 0-50 meter stations and one 0-2 meter station (27-6). The 27-6 station in the left cluster was sampled in an area where there was no sea ice present. There were no differences in community structure in the 0-50 meter depth stratum between the stations were sea ice was present and the stations were ice sea was absent (ANOSIM, R= 0.052, p = 0.392).



vegdist(dat.cluster) hclust (*, "ward.D2")

Figure 10: Cluster Dendogram of the community structure. Stations that are clustered together showed similar species composition.

From all six tested environmental parameters (Appendix 2), the distribution of the mesozooplankton species in the 0-2 meter water layer was best explained by the two environmental variables, chlorophyll *a* and sea ice ridges (Table 3).

Environmental variables	correlation	Significance
Chlorophyll a	0.5758	0.048
Chlorophyll <i>a</i> + Sea-ice ridges	0.9879	0.010
Chlorophyll a + Sea-ice ridges + Temperature	0.8061	0.036
Chlorophyll <i>a</i> + Sea-ice ridges + Temperature+ Sea-ice cover	0.8061	0.033
Chlorophyll <i>a</i> + Sea-ice ridges + Temperature+ Sea-ice cover + Salinity	0.9394	0.079
Chlorophyll <i>a</i> + Sea-ice ridges + Temperature+ Sea-ice cover + Sea-ice thickness	0.7576	0.109

Table 3: BioEnv analysis results of the 0-2 meter depth stratum stations. The significance was established by a mantel test.

There were significant differences between the δ^{13} C values in the 0-50 meter depth stratum (Oneway ANOVA, F= 5,275, df= 42, p = 0.002; Figure 11). The 4000 µm size fraction had a significant higher δ^{13} C value than the 1000 µm size fraction (Tukey HSD, p = 0.002) and the 2000 µm size fraction (Tukey HSD, p = 0.031). The 250 µm size fraction had a significant higher δ^{13} C value than the 1000 µm size fraction (Tukey HSD, p = 0.039). No significant differences were found for the δ^{15} N values (One-way ANOVA, df = 42, F = 2.087, p = 0.102).



Figure 11: δ^{13} C and δ^{15} N values for the different size fractions of the 0-50 meter depth stratum. The mean per fraction is plotted and the error bars indicate the 95% confidence interval.

There were no significant differences in the δ^{13} C (One-Way ANOVA, df = 24, F = 0.936, p = 0.464) and δ^{15} N values (Tukey HSD, p > 0.05 for all pairwise comparisons) within the 0-2 meter depth stratum (Figure 12).



Figure 12: δ^{13} C and δ^{15} N values for the different size fractions of the 0-2 meter depth stratum. The mean per fraction is plotted and the error bars indicate the 95% confidence interval.

 δ^{13} C were significantly higher in the 0-2 meter depth stratum, in comparison with the size fractions from the 0-50 meter depth stratum, for 500 µm (Independent samples t-test, df = 12, t= -2.498, p = 0.028), the 1000 µm (Independent samples t-test, df = 12, t= -2.516, p = 0.027) and the 2000 µm size fraction (Independent samples t-test, df = 11, t= -6.721, p < 0.001; Figure 13).



Figure 13: Comparison between the δ^{13} C values in the 0-2 meter depth stratum and the 0-50 meter depth stratum. The error bars represent the 95% confidence interval.

3.2. Management

3.2.1. CCAMLR

The data used for the scientific recommendations are either from national research projects of the member states or from the joint CCAMLR research programs, which consists of four working groups and one specialist subgroup (CCAMLR, 2016e) (Table 4).

Table 4	Overview	CCAMLR	working	groups	and	their	tasks
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Working Groups	Tasks
Working Group on Ecosystem Monitoring and Management	 krill stock assessment and management advice predator/ prey/ fisheries interaction physical factors which might influence the ecosystem recommends research which should be conducted for the necessity of maintaining populations responsible for CCAMLR Ecosystem Monitoring Program (CEMP) (CCAMLR, 2015c)
Working Group on Fish Stock Assessment	 assess fish stocks and give management advice recommend further research (CCAMLR, 2016f)
Working Group on Statistics, Assessments and Modelling	 evaluation and assessment of research methods identification of new research methods to establish a better scientific basis (CCAMLR, 2014)
Working Group on Incidental Mortality Associated with Fishing	 mortality of seabirds through fishing long-line fishing incidental mortality (CCAMLR, 2012b)
Sub-working Group on Acoustics, Survey and Analysis methods	 assessment and evaluation of acoustic measurements of Antarctic krill stocks (CCAMLR, 2012a)

One working group of CCAMLR is engaged in krill research and one sub-working group is solely occupied with Antarctic krill research (Table 4). The focus of the research effort on krill is based on the assumption that krill is "extremely important, because they are the main diet for most of the marine predators [...] in the Southern Ocean" (CCAMLR, 2015b). CCAMLR adopted next to its general conservation measure for all species in the convention area, nine conservation measures for the sustainable commercial exploitation of Antarctic krill (*E. superba*). CCAMLR also mentions the importance of sea ice for the life cycle of krill and the danger of melting sea ice for climate change to the ecosystem (CCAMLR, 2015b). No mesozooplankton is named in the tasks of the working groups.

To detect changes in the ecosystem caused by harvesting living resources in the convention area, the CCAMLR Ecosystem Monitoring Program (CEMP) was set up. The CEMP uses solely organisms which are dependent on the commercial fished species as health indicators for the ecosystem (CCAMLR, 2013a). The current health indicators are:

- adélie penguin (*Pygoscelis adeliae*)
- chinstrap penguin (P. antarctica)
- gentoo penguin (*P. papua*)
- macaroni penguin (*Eudyptes chrysolophus*)
- black-browed albatross (Thallasarche melanophrys)
- antarctic petrel (Thalassoica antarctica)
- cape petrel (Daption capense)
- antarctic fur seal (Arctocephalus gazella) (CCAMLR, 2013a).

As the health indicators for CEMP are high trophic levels, it could be that mesozooplankton is indirectly taken into account, if the indicator species are foraging on them. Therefore the diets of the health indicators are summarized in Table 5.

Table 5:	Diet of	СЕМР	health	indicator	species
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Species	Diet
Adélie Penguin	The diet changes with season and colony habitat. The two most dominant species in their diets are krill, especially (<i>E. crystallorophias</i> or <i>E. superba</i>), and Antarctic silverfish (<i>P. antarctica</i>) (Juáres et al. 2016; Ratcliffe & Trathan 2011; Ainley et al. 1998). The main prey of Antarctic silverfish is mesozooplankton (Pinkerton & Bradford-Grieve 2014).
Chinstrap Penguin	Chinstrap penguins are specialists, foraging especially on Antarctic krill (<i>E.superba</i>) (Niemandt et al. 2016; Ratcliffe & Trathan 2011; Rombola et al. 2010). Only occasionally they predate on fish, mostly myctophids (Ratcliffe & Trathan 2011; Rombola et al. 2010), which prey on mesozooplankton (Pinkerton & Bradford-Grieve 2014).
Gentoo Penguin	Gentoo are generalists regarding their diet (Handley & Pistorius 2016; Ratcliffe & Trathan 2011). They eat a wide range of fish, squids and crustaceans depending on the season and the colony habitat (Ratcliffe & Trathan 2011). Dependent on location, either krill species or a diverse array of fish species dominate their diet in the breeding season (Handley & Pistorius 2016; Juáres et al. 2016).
Macaroni Penguin	Macaroni penguins prey mainly on krill species and myctophids fish, depending on prey availability (Niemandt et al. 2016; Ratcliffe & Trathan 2011). Amphipods and squid also contribute to their diet in insignificant amounts in comparison with krill and myctophids fish (Ratcliffe & Trathan 2011).
Black- browed Albatross	The Black-browed albatross is a neritic forager which feeds mainly on fish, followed by squid, jelly fish and to a lesser extent crustaceans (Suazo 2008). Granadeiro et al. (2014) showed that some birds are following fishing vessels and that large part of their diet consists of discard from commercial fisheries.
Antarctic Petrel	Antarctic petrels main prey are crustaceans and fish species (Ainley et al. 1992). They are krill dependent and prey especially on <i>E. superba</i> (Nicol 1993). Yet, in pack ice they switch to a more fish based diet, mostly <i>P. antarctica</i> (Lorentsen et al. 1998; Arnould & Whitehead 1991).
Cape Petrel	The diet of Cape petrel is similar to the diet of Antarctic petrel, thus including <i>E. superba</i> and <i>P. antarctica</i> as main prey (Arnould & Whitehead 1991). However, <i>E. superba</i> constitutes a higher proportion of the diet of Cape petrel than Antarctic petrel (Arnould & Whitehead 1991).
Antarctic fur Seal	The main prey item is <i>E. superba</i> , followed by fish, squids and penguins (Casaux et al. 2016; Harrington et al. 2016). The main fish species foraged on are <i>P. antarctica</i> and myctophids dependent on the season (Harrington et al. 2016).

It is not found that the CEMP health indicators are covering all commercially harvested species, as except for the Black-browed Albatross most are dependent on krill, especially Antarctic krill (*E. superba*). Except for the Chinstrap Penguin (*P. antarctica*), all indicator species are to a greater or lesser degree generalist feeders, including fish species in their diet. The most common fish prey species are Antarctic Silverfish (*P. antarctica*) and myctophids fish species (Table 5). All of these fish species are known to forage on mesozooplankton (Ratcliffe & Trathan 2011; Ainley et al. 1998). Therefore the food path of mesozooplankton is partly covered with the CEMP health indicators. Yet, generalist species can switch from diet if certain species are scarce and are in this scene not a good health indicator. In addition, all CEMP health indicators for ecosystem changes (Hilty & Merenlender 2000).

3.2.1.1. Mesozooplankton as health indicator

It was analysed, via the criteria put forward by Hilty & Merenlender (2000), if mesozooplankton would be a good health indicator for the ecosystem of the CCAMLR convention area (Table 6). This study found that *C. acutus, Metridia* sp. and *Ctenocalunus* sp. were the most abundant species in the south-eastern Weddell Sea, therefore special attention was given to these three species. Good health indicator species do not necessary fulfil all of the criteria from Hilty & Merenlender (2000), but multiple ones.

Baseline Information	Mesozooplankton	Criterion achieved?
Clear taxonomy	The taxonomy of mesozooplankton in the Southern Ocean is widely established (Atkinson & Ward 2012). There are databases including taxonomic data and annual studies are conducted (McLeod et al. 2010).	Yes
Biology and life history	The biology and life history of the mesozooplankton in the Southern Ocean is not known for all species, but are established for <i>C. acutus</i> , <i>Metridia</i> sp. and <i>Ctenocalanus</i> sp. (Atkinson & Ward 2012; Pasternak 2001; Schnack-Schiel & Hagen 1995).	Partly Yes
Tolerance level known	There are a few studies concerning Antarctic zooplankton responses to temperature rise and ocean acidification caused by global warming (Mackey et al. 2012). However, no studies about mesozooplankton tolerance levels for stressors caused by human activities could be found.	No
Correlation to ecosystem changes established	There are several studies indicating that (meso)zooplankton size structure, abundance and community composition has a correlation with ecosystem changes (Gorokhova et al. 2016; Cairns et al. 1993; Connors et al. 1978).	Yes
Locational Information		
Cosmopolitan distribution	This criterium includes that the species are not migratory and can be found around the whole Southern Ocean (Hilty & Merenlender 2000). There are several studies that indicate the cosmopolitan distribution of important Southern Ocean Mesozooplankton such as <i>C. acutus, Metridia</i> sp. and <i>Ctenocalanus</i> sp. (Stevens et al. 2015; Atkinson & Ward 2012; Schnack-Schiel & Hagen 1995).	Yes
Limited mobility	As plankton is dependent on currents and cannot swim actively, except for vertical movement patterns, mesozooplankton cannot avoid any disturbance (Hays 2003). Therefore this criterion is fulfilled.	Yes
Niche and life history characteristics		
Early warning and functional over range of stress	Low trophic levels such as mesozooplankton, are good early warning indicators, as their community structure, abundance or size structure changes rapidly to changes in the environment (Hilty & Merenlender 2000; Connors et al. 1978).	Yes
Trends detectable	Trends are detectable and are easy to quantify as mesozooplankton is rather abundant. Yet, only long- term trends in mesozooplankton	Partly Yes

Table 6: Criteria used to identify good health indicators applied to mesozooplankton: Scale: No; Partly Yes; Yes

	communities are meaningful (McLeod et al. 2010).	
Low variability	Low trophic levels have the drawback of high variability and therefore it is hard to detect if changes occur due to natural fluctuation or due to human introduced stressors(Hilty & Merenlender 2000).	No
Specialist	Most mesozooplankton species are herbivores, however some forage on small proportions of protozoans and metazoans, thus are omnivores (Pasternak 2001). <i>C. acutus</i> is an herbivore species, while <i>Metridia</i> sp. and <i>Ctenocalanus</i> sp. are omnivores (Marrari et al. 2011a).	Partly Yes
Easy to find and measure	Sampling in the Southern Ocean is always hard to conduct, but in general mesozooplankton is not difficult to find and to measure (Atkinson & Ward 2012). It is also easier to sample compared to krill and fish since you can use smaller gear.	Yes
Other		
Taxa representing multiple agendas	Mesozooplankton is not commercially harvested nor can it be used as flagship species. Therefore mesozooplankton cannot function as indicator which is on multiple agendas.	No
Multiple indicators used	Changes in mesozooplankton community structure or abundance can indicate different natural or human introduced stressors in the ecosystem. Thus it cannot function as indicator for just one stressor (Mäkinen et al. 2016).	No

The criteria analysis results in 6=Yes, 4= No, 3= Partly Yes. This indicates that mesozooplankton can be a suitable health indicator (Hilty & Merenlender 2000). Hilty & Merenlender (2000) stated that not all health indicators criteria are equally important as it depend solely on the case. The high variability is the largest drawback of (meso)zooplankton as a health indicator and the early warning function is the biggest advantage (Gorokhova et al. 2016).

3.2.2. WSMPA

In 2002, at the World Summit on Sustainable Development, the target of the establishment of a MPA network was agreed (CCAMLR, 2016c). These MPAs should be constructed based on international law and best available scientific knowledge. In this spirit CCAMLR held a MPA workshop in 2005 which main conclusion was that a MPA network in the Southern Ocean would highly contribute to the objectives of its convention (CCAMLR, 2016c). Before that, especially fishing bans were used as a tool to protect commercially fished species in a certain season (Hawkey et al. 2013). In contrast to this single species approach, the MPA tool fits better in their ecosystem based and precautional approach of conserving the Southern Ocean resources, as it takes the whole ecosystem into account. The convention of biological diversity states that MPAs should include some, or all of the criteria in Table 7 (Hawkey et al. 2013).

Table 7: MPA criteria	(Hawkey et al. 2013)
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1.	Unique, rare or endemic species, habitats, or oceanic features
2.	Special importance for the life history stages of certain endangered or threatened species
3.	Habitats which are essential for the survival of target species
4.	High diversity, whether the diversity is within the ecosystems, habitats, communities, species or
	genetic diversity
5.	High degree of naturalness (a low level of human induced disturbance)

Mesozooplankton fits in the MPA criteria one, three and four, whereas sea ice fits in all five MPA criteria (Table 7):

- 1) Paralabidocera antarctica (Swadling et al. 2004) and Stephos longipes have adapted to live inside the sea ice matrix (Arndt & Swadling 2006b; Schnack-Schiel et al. 2008). Sea ice accumulates in the Weddell Sea as a result of convergent hydrological and meteorological conditions (Massom & Stammerjohn 2010; Brierley & Thomas 2002). In addition, perennial sea ice is rare in the Southern Ocean, and only found in the Ross Sea and Weddell Sea (Kramer et al. 2011; Arndt & Swadling 2006). As perennial sea ice contributes to species diversity and serves as refugia for sympagic organisms in summer, it is important to protect areas where perennial ice accumulates. In addition, ice-algae production in winter is crucial for juvenile organisms to overcome their first winter as lipid reserves are not yet adequate (Massom & Stammerjohn 2010).
- 2&3) Mesozooplankton is the main prey of the Antarctic silverfish (*P. antarctica*) (Pinkerton & Bradford-Grieve 2014) which is in some areas of the Southern Ocean an ecological equivalent or even substitute to the known krill based food pathway (Pinkerton & Bradford-Grieve 2014; Eastman 1985). Their protection and the protection of its habitat is therefore essential to ensure good functioning of the entire ecosystem and the survival of target species. Sea ice is a habitat for rare sympagic species and essential for target species such as *E. superba* (David et al. 2017). *E. superba* is an important food source for a lot of higher trophic levels such as the endangered Fin whale (*Balaenoptera physalus*) (Joiris & Dochy 2013).
- Sea ice provides a different habitat in the otherwise pelagic water column of the 'open sea' in the Southern Ocean which is highly diverse (Teschke et al. 2016a). In addition, Antarctic mesozooplankton is a highly diverse group, including divers Phyla of which each consists of many species of crustaceans (Boysen-Ennen et al. 1991).
- 5) Only a minor toothfish fishery is established in the South Eastern Weddell Sea (Teschke et al. 2016b). Thus it has a high degree of naturalness and is a perfect place for monitoring sea ice dependent species (Teschke et al. 2013).

In 2011 the CCAMLR adopted the "General framework for the establishment of CCAMLR Marine Protected Areas" and the convention area was split up in 9 MPA planning domains (Figure 14; CCAMLR, 2016c):



For the delineation of the MPAs in the planning domains, a GIS-based Marine Spatial Planning tool was developed to perform Multi Criteria Analysis (MCA)(Sharp & Ollivier 2012). In 2012 Germany took the lead of preparing a proposal for an MPA in the Weddell Sea (Hawkey et al. 2013). The Weddell Sea fulfils all the MPA criteria named above (Teschke et al. 2016a). Especially the 'high degree of naturalness' gives the planned Weddell Sea MPA high value concerning science, as human undisturbed areas are rare on the planet and provide scientists unique research chances (Hawkey et al. 2013). That is the reason why one of the objective for the WSMPA is "The establishment of scientific reference areas for monitoring natural variability and long-term change[...]" (Teschke et al. 2016a). Criteria for the MCA were chosen based on spatial protection of certain important features, organisms or on the basis of cost calculations (Table 8).

Subjects	Category	Criteria
Environmental	Benthic regionalisation	Bathymetry; Sedimentology (grain size, sediment distribution)
Parameters	Pelagic regionalisation	Bathymetry; Oceanography (temperature, salinity, currents), Sea-ice dynamics
	Chlorophyll- a concentration	Phytoplankton abundance
Factorizat	Pelagic ecosystems	Antarctic krill (suitable habitat, adult abundance, larva abundance); Ice krill (adult krill habitat); pelagic fish (Antarctic silverfish abundance)
Parameters	Benthic ecosystems	Zoobenthos (Shelf and Slope, Deep Sea); Demersal fish (nesting sites and suitable habitat, adult toothfish)
	Birds	Antarctic petrel foraging habitat, Breeding and non- breeding foraging habitat of the Adelie penguin, Breeding area Emperor Penguins
	Marine Mammals	Seal abundance, Whale abundance
Cost layers	Fisheries	Fishing activity, reachability of area

Table 8: Criteria used for the MCA of the Weddell Sea MPA scenario development in 20	16 (Teschke et al. 2016b)
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The sea-ice criteria, which are placed within the category pelagic regionalisation are ranked the highest in the MCA were no sea ice is detected, as it is assumed that there is more light transmission and therefore more primary production (Teschke et al. 2016b). Sea ice is taken into account for the habitat suitability criteria of *Euphausia superba* (Antarctic krill) and *Euphausia crystallorophias* (Ice krill). Yet, the importance of sea ice for certain zooplankton species as habitat and the primary production of the sea -ice algae are not taken into account. The MPA proposal also mention the importance of zooplankton for the ecosystem in their planning domain (Teschke et al. 2016b; Delegations of the USA and New Zealand 2013). The WSMPA proposal points out that the krill species in their planning domains are not so abundant and that mesozooplankton plays an important role in this ecosystem (Teschke et al. 2016b).

There are 6 General Objectives for the WSMPA (Table 9). All of them are in accordance with CCAMLR convention article II and IV (European Commision 2015).

Table 9: General objectives for the WSMPA (European Commision 2015)

1.	Protection of representative examples of pelagic and benthic ecosystems, biodiversity and habitats (including the environmental and ecological conditions supporting them) of the Weddell Sea Planning Area
2.	Protection of pelagic and benthic habitats and ecosystems which are rare, unique, vulnerable, diverse and /or endemic to the Weddell Sea Planning area
3.	Protection of areas, environmental features and species (incl. populations and life history stages) on various geographical scales which are key to the functional integrity and viability of local ecosystems and ecosystems processes in the Weddell Sea Planning Area
4.	Establishment of scientific reference areas to study, in particular representative, rare, unique and /or endemic examples of marine ecosystems, as well as biodiversity and habitats, and to monitor the effects of climate change, fishing and other human activities in the Weddell Sea Planning Area
5.	Protection of essential habitats for top predators such as marine mammals and seabirds in the Weddell Sea Planning Area
6.	Protection of essential habitats in the Weddell Sea Planning Area as potential refugia for, inter alia, top predators, fish and other ice-dependent species, in order to maintain and /or enhance their resilience and ability to adapt to the effects of climate change

Mesozooplankton fits in the objectives one, three, four and six of the general objectives of the WSMPA, whereas sea ice fits in all six criteria (Table 9):

- 1) This study found that mesozooplankton can have higher carbon availability to higher trophic levels in the Weddell sea than krill. The pelagic ecosystem food web pathways of krill and mesozooplankton are both representative examples of the pelagic ecosystem in the Weddell Sea (Boysen-Ennen et al. 1991). The Weddell Sea accumulates sea ice and has next to seasonal ice also perennial pack ice (Brierley & Thomas 2002). Therefore this habitat is a representative example of the Weddell Sea planning area ecosystem.
- 2, 3&4) Mesozooplankton is a key group for the functioning of the ecosystem in the Southern
 Ocean and especially in the Weddell Sea, where krill biomass is lower than in other parts
 of the Southern Ocean (Flores et al. 2008; Van De Putte et al. 2006; Shreeve et al. 2005;
 Pakhomov et al. 2000; Boysen-Ennen et al. 1991). Thus, it is representative for the

Weddell Sea marine ecosystem. Perennial sea ice accumulates in the Weddell sea and might become a unique feature of the Weddell Sea in the coming decades, due to the expected sea ice decline through climate change in the Southern Ocean (Massom & Stammerjohn 2010; Brierley & Thomas 2002). The sympagic mesozooplankton community as well as organisms which are dependent on sea ice in certain life history stages e.g. the Antarctic silverfish (*P.antarctica*) (Teschke et al. 2016a), are depending on the Weddell Sea sea-ice and a reference area in this region would bring unique findings.

- 5) In general sea ice is important for a lot of species in the Southern Ocean at least at one point in their life history (Teschke et al. 2016a). Thus sea ice is essential for the whole ecosystem and therefore also for the top predators. In addition, sea ice is a unique habitat for several top predators in the Weddell Sea. All penguins depend on the sea ice, especially the emperor penguin which breeds and raises his offspring on the sea ice (Jenouvrier et al. 2014). Also the six 'true Antarctic species' of seals depend on sea ice and are all present in the Weddell Sea (Teschke et al. 2016a).
- 6) Sea ice and especially perennial sea- ice, which is accumulating in the Weddell sea (Massom & Stammerjohn 2010), is a unique and essential habitat for lower trophic levels as well as for top predators (Teschke et al. 2016a). Sympagic mesozooplankton finds refuge and food, in form of ice-algae, in the sea ice channels (Jia et al. 2016; Arndt & Swadling 2006; Brierley & Thomas 2002; Schnack-Schiel et al. 2001). The Weddell Sea ice might be one of the only sea ice refuge areas in the Southern Ocean in the future and could play a crucial role in slow adaption of the ecosystem to the climate change (Massom & Stammerjohn 2010).

4. Discussion

This study tried to answer the question how important mesozooplankton is, including its relation to sea-ice, in the food web of the south-eastern Weddell Sea. These ecological findings were than used, in combination with literature study, to state the implications for future management decisions of CCAMLR and the WSMPA project team.

4.1. Ecology

Mesozooplankton contributed significantly more to the carrying capacity of the south-eastern Weddell Sea than krill. The relative as well as the total biomass of mesozooplankton was higher than krill biomass in the 0-50 meter depth stratum. The combined total biomass of mesozooplankton and krill was higher in the 0-50 meter depth layer in comparison to the 0-2 meter depth layer. The biomass of krill did not differ between the two depth strata, but mesozooplankton biomass was significantly higher in the 0-50 meter depth stratum in comparison to the 0-2 meter depth stratum. This finding complements the results of Flores et al. (2008); Van De Putte et al. (2006); Pakhomov et al. (2000) and Boysen-Ennen et al. (1991) which all indicate that mesozooplankton is at least as important as krill in the food web of the Southern Ocean.

All 0-50 meter depth stratum stations had higher relative biomass of mesozooplankton than krill, except for station 43-1, where half of the relative biomass was composed of krill. This station was the only shelf sampled station and the findings are in accordance with the results of Shreeve et al. (2005) which found that krill biomass is highest at the shelf regions.

In the 0-2 meter depth stratum, biomass was overall very low at stations with higher krill biomass. This could be a result from competition over phytoplankton as food source or due to predation of krill on mesozooplankton (Marrari et al. 2011b). Krill forages over three trophic levels. Most of their diet consists of phytoplankton, followed by protozoans and in smaller amounts mesozooplankton, mainly copepods (Atkinson et al. 2012). During the sampling of this study the chlorophyll *a* values were < 0.5 mg m⁻³ where krill biomass was high (Appendix 2), which is considered low (Teschke et al. 2016b) and which could be an indication for high predation.

In the summer in the south-eastern Weddell Sea, *C. acutus* could be the primary carbon vector, constituting the most to the carrying capacity of the ecosystem. This was indicated by the high relative biomass in the 1000 μ m size in the 0- 50 meter depth stratum. This corresponded with high abundances of *C. acutus*, which was the most abundant copepod and mesozooplankton species during this study (39 % over all stations) and a major constituent of the 1000 μ m size fraction. In the winter *C. acutus* overwinters in diapause at depth and migrates at the beginning of spring to the surface layers, where it exploits the phytoplankton bloom (Schnack-Schiel & Hagen 1995). Hence, in winter *C. acutus* is absent from the surface layers and a shift is seen towards smaller size fractions, mostly dominated by *S. longipes* and Ctenocalanus *sp.* (David et al. 2017).

The C:N ratio of all size fractions and depth strata indicated that in general mesozooplankton was in a an good body condition, with a value around 4.5 (Harris et al. 1986), and therefore a good food source for higher trophic levels. As the samples were taken at the beginning of summer, mesozooplankton had probably already started to accumulate their lipid reserves for the next winter (Shreeve et al. 2005; Schnack-Schiel & Hagen 1995). The C:N ratios for the lower size fractions (250

 μ m and 500 μ m) in the 0-2 meter depth strata were significantly higher in comparison to the C:N ratio of the other size fractions of this depth stratum. The smaller sympagic organisms are able to retreat into the sea-ice matrix (Kramer et al. 2011; Kiko et al. 2008; Schnack-Schiel et al. 2004; Schnack-Schiel et al. 2001), this could give them access to a food source, namely sea-ice algae, that is hidden from the larger mesozooplankton, which may results in higher C:N ratios. If so, results suggest that sea-ice algae are potentially a good quality food source for small mesozooplankton.

The 0-2 meter depth stratum with sea ice clearly houses a community that is different from the 0-2 meter open water layer and the 0-50 meter water column. The mesozooplankton species composition was significantly different in the 0-2 meter depth stratum comparison to the 0-50 meter depth layer. It consisted for 81% of copepods in the 0-50 meter depth stratum and 94% in the 0-2 meter depth stratum. The 0-50 meter depth layer samples and the one 0-2 meter depth layer sample without sea-ice were dominated by *C. acutus* which is a typical species encountered in surface waters at this time of the year (Shreeve et al. 2005; Schnack-Schiel & Hagen 1995). The species encountered in the 0-2 meter depth stratum at the stations where sea ice was present were dominated by *P. antarctica*, a copepod that is associated with the sea ice and is considered a sympagic species (Swadling et al. 2004). As *C. acutus* belongs in the 1000 µm size fraction and *P. antarctica* in the 500 µm size fraction, this finding indicates that the 0-2 meter depth layer community structure and size structure is only significant different from the 0-50 meter depth layer if sea ice is present.

This is verified through the results of the cluster analysis, as stations in the 0-2 meter depth stratum with sea ice present were clustered together, while station 27-6, which lacked sea ice, was found in the cluster containing samples of the 0-50 meter depth stratum. Interestingly, this study did not find high abundances of *S. longipes* in the 0-2 meter depth stratum which is often found to be dominant in the Weddell Sea in the surface waters in summer, where sea ice is present (Kramer et al. 2011; Schnack-Schiel et al. 2008b; Arndt & Swadling 2006; Schnack-Schiel et al. 2004). *P. antarctica* and *S. longipes* rarely co-occur in high numbers (Arndt & Swadling 2006) and it seems that *P. antarctica* successfully outcompeted *S. longipes* in the summer of 2014/2015.

Sea ice seemed to have a contrasting effect on the biomass found in both depth strata. In the 0-2 meter depth stratum, biomass was very low at the stations with sea ice, but high at station 27-6 were sea ice was absent. In the 0-50 meter depth stratum biomass was higher at stations were sea ice was present. This is potentially a result of the low chlorophyll *a* values (< 0.5 mg m⁻³) at stations in the 0-2 meter water layer where sea ice was present (Appendix 2; Teschke et al. 2016b), which suggests that primary production was low at the ice-water interface. Higher chlorophyll *a* values were found at the open water station in the 0-2 meter water layer (Appendix 2). Chlorophyll *a* content resembles the primary production and often corresponds with high biomass in an area (Wallis et al. 2016).Therefore it can be assumed that there were higher chlorophyll *a* concentrations in the 0-50 meter depth stratum. However, as environmental variables were missing for the 0-50 meter depth stratum, it could only be hypothesized why there was such high mesozooplankton biomass.

In the 0-2 meter depth stratum, the species distribution was best explained by the chlorophyll *a* content and the presence or absence of ridges under the sea ice. Ridges underneath the sea ice can be hotspots of algal aggregations, and provide shelter from currents (Katlein et al. 2015). Thus, at the sea-ice stations mesozooplankton biomass and abundance could actually have been high where ice-algae accumulate in sea-ice ridges and low or absent where no ridges were present (Katlein et al.

2015). This could result in the overall low abundance and biomass of mesozooplankton at the sea-ice stations in the 0-2 meter depth stratum, reflecting a patchy distribution of mesozooplankton that was generalised over the sampling transect. In addition, summer ice is more porous and more habitable space is present than in winter (Arrigo 2014). Densities of copepods in rotten summer ice can reach up to 1000 individuals L^{-1} (Schnack-Schiel et al. 2008). Thus sympagic copepods could have migrated into the perennial sea ice matrix, leaving lower abundances of mesozooplankton in the water layer directly underneath the sea ice.

The significant higher δ^{13} C of the mesozooplankton from the 250 and 500 µm size fraction in the 0-50 meter depth stratum indicate that these size fractions were relying more on ice-algal produced carbon that on pelagic phytoplankton produced carbon (Kohlbach et al. 2016). In the 0-2 meter depth stratum, the absence of differences in the δ^{13} C values suggest that the organisms present at the ice-water interface utilised the same food source. In addition, in the 0-2 meter depth stratum all δ^{13} C values were significantly higher for all size fractions in comparison to the 0-50 meter depth stratum, except for the 4000 size fraction, which is macrozooplankton. This indicates that the organisms under the sea ice were more dependent on ice-algal produced carbon in comparison with the organisms in the 0-50 meter depth stratum (Kohlbach et al. 2016). Thus, even in summer when the samples for this study where taken, carbon produced within the sea ice serves as food for organism that reside under the ice-water interface. In the 0-50 meter depth stratum, pelagic production probably plays a more important role.

In this study, differences in δ^{15} N values per size fraction were not found, thus no trophic level differences between the size fractions in both the 0-2 and 0-50 meter depth strata were present. *C. acutus* is a grazer, while *C. propinquus* and *Metridia sp.* are considered to be omnivores (Atkinson et al. 2001; Schnack-Schiel & Hagen 1995). In addition, species of the genus *Paraeucheata* are predators (Atkinson et al. 2001). Thus, several trophic levels were present inside each size fraction, which probably made it impossible to detect different trophic levels between the size fractions.

4.2. Management

The general perspective on the Southern Ocean ecosystem is that this area is a krill dominated system (Brierley & Thomas 2002; Atkinson et al. 2001). However, our results show that in the south-eastern Weddell Sea mesozooplankton, especially copepods are dominant over krill. This indicates that mesoozplankton is an important vector for carbon in the south-eastern Weddell Sea during the summer season. This confirms indications from previous research that the Weddell sea is a copepod dominated ecosystem (Van De Putte et al. 2006; Pakhomov et al. 2000). In addition, even in summer ice-algal produced carbon seemed to be important for organisms that resided under the sea ice (0-2 meter) and the sea-ice habitat hosted a specific mesozooplankton community, stressing the importance of sea ice for this food web. Therefore this study recommends taking mesozooplankton and sea ice into account for the management over the Weddell Sea /Southern Ocean.

The ecosystem health indicators CCAMLR used to assess ecosystem changes are seals and birds, which are mostly krill dependent (Table 5). These indicators are appropriate in regions were krill is the primary carbon vector, but probably not in regions were mesozooplankton fulfils this role. In addition, the indicator species are chosen based on their dependency on commercial fished species and focus completely on top-down control (CCAMLR, 2013a). Therefore, the CEMP health indicator

species are unable to serve as early warning indicators, as higher trophic levels show a delayed or weakened response to changes in the ecosystem (Hemraj et al. 2017; Gorokhova et al. 2016). (Meso)zooplankton can fulfil this function as early warning indicator of change in marine ecosystems (Gorokhova et al. 2016). This way CCAMLR could cover a larger range of the ecosystem and the ecosystem assessment would become a two-way process, by covering the ecosystem with bottom-up and top-down health indicators (Gorokhova et al. 2016).

Changes in (meso)zooplankton community structure, abundance, biomass or size structure can give early indications that the ecosystem is shifting (Gorokhova et al. 2016; Guan et al. 2011). In the past, especially fresh water- and coastal lagoons studies, focused on zooplankton as health indictors (Blank et al. 2017; Hemraj et al. 2017; Gorokhova et al. 2016; Guan et al. 2011). Setting up a good zooplankton health indicator in the marine environment is harder, due to the bigger scale of the environment and the variability of zooplankton in this large area (Gorokhova et al. 2016). Yet, the importance of zooplankton for the marine ecosystem and therefore for ecosystem based management is recognized (European Commission, 2017); Gorokhova et al. 2016; Teschke et al. 2016b).

Taxonomic composition can be very informative in terms of ecosystem assessment, yet can only be used in a small spatial scale as natural variability in marine ecosystems is high and dominant taxa alternate per region (Gorokhova et al. 2016). In addition, taxonomic studies are rather time consuming and therefore expensive. However, a size based approach can be used. Gorokova et al. (2016) found that mean size of zooplankton in combination with biomass or abundance data was the most reliable way of indicating changes in the ecosystem of the Baltic Sea (Gorokhova et al. 2016).

Changes in size spectra of zooplankton community can impact the dynamics of the whole marine ecosystem (Connors et al. 1978). This study found that the 1000 μ m size fraction was most abundant in the summer in the south-eastern Weddell Sea. If this size composition permanently shifts to a smaller or larger fraction over several years, it indicates changes in the ecosystem (Gorokhova et al. 2016; Guan et al. 2011). Larger zooplankton are considered better food for zooplanktivorous organisms as less prey items have to be consumed to meet their food demand (Gorokhova et al. 2016). In addition to the size class, biomass or abundance of (meso)zooplankton has to be measured as well as it could be that the size fraction distribution remains the same, but there is a drop in total biomass (Gorokhova et al. 2016).

The Southern Ocean is a diverse area and instead of developing a new framework to incorporate mesozooplankton in the general management of the Southern Ocean, a more regional approach would fit better. The establishment of MPA's can be used to incorporate region specific health indicators into the management of the Southern Ocean. In case of the Weddell Sea, the WSMPA can be used to serve as refuge for sea ice dependent biota and monitoring of (meso)zooplankton can be performed within this area. This way the WSMPA could serve as a reference area for a mesozooplankton food web dominated ecosystem and for the sea-ice associated communities.

In the delineation of the WSMPA, sea ice is not taken into account as a criterion itself. Sea ice and its unique features as a habitat, refugia and place for high quality food, fits in all six of the general objectives of the WSMPA and in the all five criteria for the establishment of MPAs. Therefore sea ice should be considered as criterion in the multi criteria analysis for the WSMPA establishment.

Due to the high variability of mesozooplankton distribution and abundance, it is suggested not to take zooplankton into account with the establishment of the WSMPA, but purely for the monitoring and as health indicator when the WSMPA is established.

An elemental step for setting up a health indicator is the establishment of reference conditions. A preferred way is using communities from a reference area in pristine state or historical data from a time where influence through anthropogenic stressors were low (Gorokhova et al. 2016). CCAMLR could use both methods to assess the best reference conditions. Zooplankton studies and the zooplankton database of SCARs 'Southern Ocean Continuous Plankton Recorder Surveys', is available to CCAMLR, and goes back to 30 years (McLeod et al. 2010). In addition could the WSMPA, when established, be a pristine reference area, as human activities would be down to purely scientific research.

5. Conclusion

This study found that in early summer mesozooplankton had a higher contribution than krill to the carrying capacity of the food web in the south-eastern Weddell Sea. The highest biomass was found in the 0-50 meter water layer. Yet, mesozooplankton from both depth strata had a very high C:N ratio, thus are good food sources for higher trophic levels. Mesozooplankton could function as early warning health indicator for CCAMLRs ecosystem monitoring, as mesozooplankton is low in the food chain and sensitive to environmental changes. Therefore CCAMLR should incorporate mesozooplankton in the assessment of the ecosystem. However, mesozooplankton community structure varies greatly across large geographic regions, such as the Southern Ocean. Therefore, a size-class based approach in combination with biomass or abundance data of mesozooplankton is suggested, to assess the health of the Southern Ocean. The size-based approach is less time consuming than determining the taxonomic composition, but still is very informative in terms of ecosystem functioning and allows for the identification of change in an early stage. Yet, instead of focusing on a new framework for the whole Southern Ocean, CCALMR should take the opportunity of the MPA developments. Region specific indicators, that resemble the region's specific situation, can be developed, to monitor e.g. the Weddell Sea. As sea ice seems important, both as habitat and as a place where food for higher trophic levels is produced, it is suggested that sea-ice coverage and composition should be taken into account as criteria for the WSMPA area establishment. Due to the high variability in mesozooplankton abundance, this study does not suggest to take mesozooplanton into account in the delineation of the WSMPA. However as said before, once established the WSMPA could be monitored for mesozooplankton to detect changes in the environment and the carrying capacity of the ecosystem. The WSMPA could then be used as the first reference area in the Southern Ocean which monitors pelagic mesozooplankton community structure and the sea-ice associated mesozooplankton community.

6. Recommendations for further research

- This study focused on the 0-2 and 0-50 meter depth strata. A follow up study, including also deeper water layers, would strengthen the findings and show if mesozooplankton still has the higher biomass in comparison with krill.
- Krill lives in the upper water layer of the Southern Ocean the whole year, while many copepod species, such as *C. acutus*, migrate to deeper water to overwinter (Marrari et al. 2011; Shreeve et al. 2005). David et al. (2017) found that in the winter of 2013, in the Weddell Sea, in the 0-2 meter water column copepods were numerically dominant, but most of the biomass was constituted by krill. Further studies should be conducted to investigate the inter-annual variability in the distribution and abundance of mesozooplankton and krill.
- The C:N ratios of mesozooplankton and krill could be compared in the south-eastern Weddell Sea, to get an indication which group is better quality food, in terms of lipid content.
- Research on open water where the sea ice just has retreated is missing. Therefore, it remains unknown what happens to the sympagic fauna and if there are shifts in species composition. This study recommends sampling in open water in winter and in summer, as this can give clues to how this system might be responding when sea ice decreases in the future.
- The variability of the zooplankton is mostly established by comparing research studies over a few years between seasons. In addition, different geographic areas are surveyed. To able to determine the variation correctly repeated sampling at the same stations, in the same season, should be performed.
- The copepod *P. antarctica* is usually dominant in the Indian sector of the Southern Ocean (Arndt & Swadling 2006; Swadling et al. 2004), but this study found high abundances of *P. Antarctica* in the Weddell Sea. It would be interesting to investigate if its abundance in the Weddell Sea is due to normal inter-annual fluctuations, a result of a shift in community structure, or an expansion event of the geographical range of this species.
- In this study, a net of 0.33 mm and 0.3 mm net was used to sample the zooplankton. A future study, including a smaller 150 µm net and a larger 4 mm net would allow for sampling a greater size range. Including not only mesozooplankton, but also microzooplankton and macrozooplankton in the evaluation of the carrying capacity of the Southern Ocean, would better include the complete ecosystem. These results could also be used for the development of a health indicator size-based metrics for CCAMLR, which includes not only mesozooplankton, but the whole zooplankton community.
- If a study like this one is conducted again, it would greatly improve the results if the dominant species within each size fraction were separated on board and stored separately for C:N ratio and BSI analysis. This would enhance the resolution of the data and would reveal much more insight into zooplankton throphodynamics.

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Appendices

Appendix 1:	Overview ove	er all sample	stations	used in t	this study.
Appendix 1.	010101010000	i un sumpic	. stations	ascam	uns scaay.

Sample ID	Station	Haul	Net	Depth (m)	Region	Sea Ice	Latitude	Longitude
PS89_0162	27-5	3	RMT (1-3)	0-50	Sea Ice Zone	NO	-64.0400000	-000.0700000
PS89_0195	29-3	4	RMT (1-3)	0-50	Sea Ice Zone	YES	-66.0200000	000.0500000
PS89_0239	30-2	5	RMT (1-3)	0-50	Sea Ice Zone	YES	-66.4500000	-000.0600000
PS89_0553	41-1	6	RMT (1-3)	0-50	Shelf	NO	-70.5300000	-007.8900000
PS89_0606	43-1	7	RMT (1-3)	0-50	Shelf	NO	-70.4600000	-008.3100000
PS89_0909	62-2	11	RMT (1-3)	0-50	Sea Ice Zone	YES	-69.4700000	-010.4400000
PS89_1002	66-5	12	RMT (1-3)	0-50	Open Water	NO	-69.0200000	-006.9400000
PS89_1005	70-1	13	RMT (1-3)	0-50	Sea Ice Zone	YES	-68.2500000	-003.9500000
PS89_1256	79-1	14	RMT (1-3)	0-50	Open Water	NO	-65.8300000	000.0500000
PS89_0085	24-2	2	SUIT	0-2	Sea Ice Zone	YES	-61.9850000	000.0300000
PS89_0439	27-6	3	SUIT	0-2	Sea Ice Zone	NO	-64.1100000	-000.0460000
PS89_0070	29-1	4	SUIT	0-2	Sea Ice Zone	YES	-65.9480000	-000.0400000
PS89_0306	30-4	5	SUIT	0-2	Sea Ice Zone	YES	-66.4920000	000.0480000
PS89_1146	71-1	17	SUIT	0-2	Sea Ice Zone	YES	-68.2030000	-003.6890000

Station	Depth (m)	Chlorophyll a (mg m ⁻³)	Temperature (°C)	Salinity (ppt)	Sea-ice cover (%)	Sea-ice thickness (m)	Sea-ice ridges (#)
24-2	0-2	0.449	-1.786	33.435	83	1.02	6
27-6	0-2	2.857	-0.941	33.509	83	0	0
29-1	0-2	0.629	-1.744	33.624	79	0.71	4
30-4	0-2	0.268	-1.511	33.754	79	0.77	8
71-1	0-2	0.332	-1.526	33.627	99	1.53	20

Appendix 2: Overview over the environmental parameters measured with the CTD on the SUIT.