

REPLIES TO REVIEWER 1

We thank the reviewer for carefully reading the manuscript and providing useful comments.

5 This manuscript presents the vertical distribution and migration of macro-zooplankton in the open Cretan Sea as derived from a 75 kHz ADCP dataset that covers a period of 30 months with four deployments. The topic falls within the scope of Ocean Science and might constitute a valuable contribution to the Special Issue “Coastal marine infrastructure in support of monitoring, science, and policy strategies”. The use of ADCP as a non-invasive method to infer the zooplankton distribution has been demonstrated by
10 previous papers published on the subject in other oceanographic regions. The novelty of this study is that it applies the ADCP method for this purpose for the first time in the Eastern Mediterranean, in particular, in the Cretan Sea, a very dynamic and crucial area for the circulation of the EMed. Unfortunately, the first three ADCP deployments were shallower (down to 300 m depth) than the fourth one (450 m) and missed some interesting zooplankton features. Despite the above mentioned positive aspects, the
15 manuscript is not clear and focused enough to meet the Ocean Science standards. The manuscript should be more neat and concise to gain in clarity and needs a profound revision to solve the numerous problems I see in the various sections and have detailed in my specific comments. The main limit of this work is the lack of macro-zooplankton data collected with vertical tows that could have helped substantially to interpret the ADCP profiles. The experimental set up was clearly designed on the current measurements
20 and not on zooplankton analysis, which was decided successively (at least, this appears from the content of this manuscript). Other papers have analysed ADCP data for inferring on zooplankton vertical migration without parallel zooplankton sampling, but this does not justify the critical limit determined by this choice.

25 SPECIFIC COMMENTS

Introduction needs to be carefully revised because the issues presented are not well linked and not clear in some parts. Topics like: the biological pump, the role of zoo- plankton vertical migration, the Eastern Mediterranean and the Cretan Sea are not present adequately. The
30 biological pump should be introduced in the first sentence and expanded in the second one. The primary production is not linked to the following paragraph and can be removed because not developed further in this section. To be precise, the biological data from midwater are not lacking but they are few in comparison with epipelagic layers (L22-27, pg.1). The vertical migration of zooplankton comes out of the blue (L30); this topic relevant for this study should
35 be properly introduced and the related papers in the Med should be cited. After that, it should be provided the info on zoo vertical migration available for the EMed and explained the “different migrating strategies” emerged from previous papers. The last part of pg. 1 is quite confused and should be rewritten. The Cretan Sea should be presented in a clear way; it is necessary to provide a brief description of the Sea with basic info on characteristics of hydrology,
40 biochemistry and zooplankton in the area before claiming that it is representative of a wider EMed area.

Reply: The first part of the introduction was restructured in the following order: brief description of biological pump (without mention of primary production), diel vertical migration (DVM) description (including patterns, related factors), DVM studies by ADCP, DVM studies in the
45 Western and Eastern Mediterranean Sea (including migrating strategies), description of Cretan Sea (hydrology, biochemistry and zooplankton), Mediterranean DVM studies by ADCP. We specified the gaps of knowledge (instead of gap of data) regarding midwater depths. New references were added to support the above description.
50 The reasons why the Cretan Sea is representative of a wider area was more developed.

The second part of Introduction looks better and requires minor changes, apart the two following ones. 1-The study by Cardini et al. (2003) is misplaced here, in my opinion. If the present study has been stimulated by the hypothesis by Cardini et al. (same place, same ADCP) on zooplankton vertical migration, the rationale and the aim of the present study should be presented first and Cardini after (e.g., this study aimed at. . . .testing the hypothesis by . . .). If the two studies were not related in any case, the citation should be removed from Introduction and used in Discussion.

Reply: The two studies were related. The last paragraph of the Introduction section was rewritten to reflect this fact as suggested.

The last sentence on carbon cycle should be removed: considerations on this topic must be treated with caution and not here, because the present study does not provide results on carbon data.

Reply : The sentence was removed from the text.

Methods

The experimental set up was not designed to properly interpreting the ADCP data in relation to macro zooplankton distribution because of the lack of parallel zoo plankton sampling. The zooplankton sampling done with a 200 μm net at monthly frequency in the upper 100 m and the single oblique tow performed with a Bongo net (330 μm and 500 μm) in Dec 2013 in the 0-500 m layer were useless for interpreting the ADCP data of this work and, in fact, those zooplankton data are not presented. Therefore, this part (pg5, L7-14) should be removed.

Reply: The paragraph “At the E1-M3A zooplankton... .. mentioned briefly in the discussion.” was removed.

Similarly, the downward looking 400kHz ADCP data were not used for the present work, so this part should be removed too (pg4, L34-36).

Reply: The 400kHz ADCP data were used in Figure 3, b. The data are presented at pg. 6 L26 and pg.7 L2-4.

The sampling and analytical methods for chlorophyll concentrations in Figure 4 are not presented.

Reply: The chlorophyll concentrations were measured with the WETLABS ECO FLNTU sensors which were mounted on the profiling 19plus and moored 16plus CTDs. The above information was added to the text.

I do not understand how the burst velocity may help identifying the optimal sampling strategy for zooplankton; probably you mean “the optimal cell extension for the most appropriate recording of zooplankton signals”? Or something else? This part must be clarified (pg.3, L16).

Reply: The burst velocity was not properly defined in the text. The paragraph containing the burst velocity definition was rephrased as follows:

“One parameter used to potentially identify the optimal cell extension and sampling interval for the most appropriate recording of zooplankton signals was the hereafter defined burst speed. The burst speeds of each cell are defined as the highest and lowest vertical velocity measurements respectively during a time period of one day. The velocity measurement inside a cell over the sampling interval is the result of the averaging of several pings. As the sampling interval increases and/or the cell extension

decreases and/or the actual zooplankton speed increases, we expect the actual zooplankton speed to be underestimated because zooplankton will not be inside the cell throughout the duration of the measurement but only during a fraction of it. The largest underestimation is expected when the actual zooplankton migrating speed is maximum. Thus, comparison of upward and downward burst velocities between deployments at depths around 250 m were used to identify the most appropriate sampling scheme.”

The affirmation on pg. 4, L3 is based on statistical test? Expand the explanation.

Reply: The comment refers to the phrase “The range of the cells used (10 – 20 m) did not affect the burst velocity and the average velocity measurements.”

The sampling rate of deployments No 1, 3 and 4 was the same (30 min). If a bias was caused to the burst and average velocities due to the cell extension (as explained in reply to the previous comment) then choosing a smaller cell extension should result in smaller burst and average velocities. Smaller cell extensions of deployments with the same sampling interval did not result in smaller velocities, although the conclusion was based on visual inspection of burst and average velocities plots and not on a statistical test.

The following phrase was added to the text:

“The range of the cells used (10 – 20 m), on the other hand, did not affect the burst speed and the average velocity measurements. Based on visual inspection, smaller cell extension during the 1st, 3rd and 4th deployments (30 min sampling interval) did not result in smaller burst and average velocities.”

The Results section needs a substantial revision and a more synthetic rewriting.

The results are interesting, but their too long, detailed and sometimes repetitive description leads to lose the focus and weaken the main messages. This section is definitely too long (9 pages) and repetitive and I found it heavy to read.

Reply: Results section was reduced by about one page, some parts were rewritten, and repetitions were removed.

It is presented in subsections, but it is not clear enough because the topics cross different sections, the figures are continuously cited back and forth (not in the numerical order for figs 7,9,10,8) and this creates confusion and tiredness in the reader.

Reply: The back and forth citation of figures was reduced to a minimum. Figures’ citations are now in numerical order.

Links among figures are commented and interpretations are mixed with mere objective results. Links, interpretation, and comments should go all in Discussion to provide the reader with a neat and clear overview of the study. I suggest to follow the typical simple way of result presentation, i.e. describing objectively the data showed by the figures, in the right succession, without anticipating interpretation. This is not a “Results and Discussion” section.

Reply: Interpretations, links among figures and their comments were removed from the Results section.

Part 3.1 on Environmental conditions is confusing; it is not clear what are the general typical conditions of the area as emerged from previous works and what are the results from the present work. There should be a separated section, placed in Methods, reporting the general characteristics of the area.

Reply: A “Hydrology of the Cretan sea” subsection was added under the “Methodology” section. No references are now present in Part 3.1 of the manuscript.

The use of the past tense would help clarifying what are the results of the present study.

Reply: The results were written in past tense to clarify the results.

The “constant presence of a deep layer of scatterers” (pg8, L11) was actually recorded only in the fourth deployment because deeper down to 450 m, so it cannot be defined “constant”.

Reply: The term “constant” was removed from the text.

The affirmation that zooplankton feed at certain depths and hours (e.g. pg.9, L8 and somewhere else) should be changes in likely supposition; the present study does not demonstrate any feeding activity, which might only be hypothesized as (one of the) possible explanation to the vertical zooplankton displacement inferred by ADCP backscatter.

Reply: Changed in the text as suggested.

There are other strict affirmations that should be changed in interpretative suppositions/hypotheses (e.g.,pg.10, L7-12). Many of the migrant macro-zooplankton animals are carnivorous. This kind of interpretation should be presented with more caution.

Reply: The last paragraph of section 3.2 was removed from the manuscript and rewritten in a less affirmative way as suggested by the reviewer

Discussion

contains some repetitions of Results; the two sections should be rewritten in parallel to separate objective results from interpretation and discussion. This would make the manuscript much clearer and more interesting and pleasant to read.

Reply: Repetition of results were removed from Discussion section and parts of it were rewritten.

Discussion should be also organized in sub-sections to address the different aspects of this study.

Reply: The Discussion section is now organised in sub-sections.

Light is interpreted as “the main factor affecting zooplankton migration” (pg.14, L28); it can be a triggering signal, a factor acting directly or indirectly on individual animals, or on swarms. This interpretation should be more exhaustively expanded by using information from the literature on light perception by zooplankton groups.

Reply: The interpretation was expanded using information from the literature as suggested.

The last part of Discussion (pg.17, from L17) is dedicated to comments on the limits of this work, which are quite heavy indeed and should have been addressed at the beginning of the study, planning a more appropriate in-field-experimental design. It is right and honest to discuss on the limits, but this section should be closed with positive conclusions highlighting how this work contributes to increase the knowledge on zooplankton in the Cretan Sea. I am positive that this work might be useful to Mediterranean zoo planktonologists, but the authors should convince better the readers.

Reply: The paragraph explaining the limitations of the methodology was moved to the last part of the Methods Section under the Limitations subsection.

Technical corrections

The past tense must be used for presenting the results of this study, not the present tense.

Reply: The results were written in past tense to clarify the results.

Zooplankton, as collective name, need plural verb.

Reply: Corrected to plural.

Population must be removed or replaced by “assemblages” when associated to zoo plankton because “population” has to be used only referring to species.

Reply: Population was replaced by assemblages.

“Sampling” should be replaced by “recording” in relation to ADCP data; sampling is properly used for collection of samples (e.g. pg.3, L19-24 and somewhere else)

Reply: “Sampling” was replaced “recording” as suggested.

Some units are often written incorrectly with missing space throughout the manuscript (e.g., ms-1, cms-1 instead of the correct m s-1, cm s-1).

Reply: Units were corrected.

Velocity and speed are used indifferently (e.g. in the caption of Fig. 8); better using one of the two throughout the whole manuscript.

Reply: Speed was changed to velocity in the caption of Fig. 8 as it was used incorrectly. For the rest of the manuscript, speed was used to indicate the absolute vertical velocity or the magnitude of horizontal velocity (currents and wind).

Pg.1, L28: Basin instead of “Part” Pg2, L4: “several meters” length pertains to long chains of gelatinous zooplankton like salps, for example; it is written wrong here because it seems that some medusae (jellyfish) are several meters large, that’s not true in the Med.

Reply: “Part” was replaced by “basin”. “several meters” was removed as the whole introduction was rewritten.

Pg2, L10: delete “However” L10-13: the two sentences repeat the same concept (ADCP detects zooplankton) and should be merged.

Reply: “However” was deleted.

L35: “south Aegean Sea” must be indicated the first time the Cretan Sea is mentioned in Introduction, not here.

Reply: “south Aegean Sea” was moved to the abstract.

L37: “Water-plankton sampling” is an awkward expression; it should be Niskin sampling for chlorophyll measurements (or phytoplankton, microzooplankton) or net tows for zooplankton sampling.

Reply: The sentence was replaced by “The observing effort is complemented by monthly R/V cruises to perform CTD casts (temperature, salinity, fluorescence) and net tows (zooplankton).”

L39: 6 months not 7, from 15/11/2012 till 20/5/2015

Reply: Corrected to “six months”.

Pg3, L16: “behaviour” can be many things, so specify “vertical movement” or migration

Reply: The whole paragraph was rephrased.

Pg4, L18, L20: “pieces” should be replaced by “datasets” or “sections” or a more appropriate term.

Reply: “Pieces” was replaced by “datasets”.

L19: it’s not really a long-term, better specify seasonal and interannual variability

Reply: “long-term” replaced by “seasonal and interannual”.

L33, 34: add “depth” after 100 m and 250 m (check throughout the manuscript)

Reply: “depth” added throughout the manuscript.

Pg5, L1,2: “M” must be replaced by “months “

Reply: “M” replaced by months.

Pg6, L21: “typical values” are averages, medians? Clarify

Reply: “typical values...” replaced by “average wind stress is 0.82 Pa”

L25: zooplankton has not been introduced yet; this link between results should be moved to Discussion.

Reply: Removed as suggested.

Pg7, L5; “Figure” is repeated

Reply: “Figure” was removed.

L7: as reported by Cardin et al. (2003).

Reply: Rephrased as suggested.

Pg8, L9: . . .four ADCP deployments. . .

Reply: Rephrased as suggested.

Pg9, L10: the daily data are embedded in the graphs of Fig.5 but the daily resolution is not visible.

Reply: The “... a diel...” was removed.

Pg10, L8: might be due, not “must”

Reply: Rephrased as suggested.

Pg14, L26: I do not understand how groups A and B “are coupled in a behavioural relation”; it should be rephrased and clarified.

Reply: The paragraph containing the phrase “are coupled in a behavioural relation” was removed from the manuscript.

Figures

Fig.2- the research vessels should not be mentioned in the figure captions; the upper values of y-axis in panels a, d, e, should be indicated.

Reply: The research vessels were removed from the caption. The upper and lower values of the y-axis in panels a, d, e are now indicated.

Fig.3-the unit “m” is missing on the y-axis

Reply: Depth [m] was added to the y-axis.

Fig.4- The “grey dotted lines” are barely visible in the upper panel. I see only a single value in blue, and not a range above each cast; is it the max value?

Reply: The “grey dotted lines” was changed to “grey lines”. The light grey color for the minimum cast value was chosen because otherwise the figure would be cluttered, and the normalized chlorophyll data would not be easily visible. Above each cast there are two numbers: the original chlorophyll range of the cast (in blue) and the original minimum chlorophyll value (in red). The words “Min” and “Range”, colored red and blue respectively, are shown to the left of the colored numbers.

Fig.5- The explanation of the black and yellow circles in the caption is wrong, it’s the opposite. Repeat here the explanation of the triangles and bars.

Reply: The explanation of the black and yellow circles was corrected, and the explanation of the triangles and bars was added to the caption.

Fig.6- The day and night times should be indicated in this figure as referred to in the text.

Reply: The day and night times were added to the figure.

Fig.8- I do not see the 4 hours indicated in the text (pg.10, L26); the hours should be indicated in the graph. I suppose that “largest speed measurements” mean actually “highest speed values”(or velocity?).

Reply: The hours indicated in the text were added to the figure. The caption was rephrased to “(a) Instantaneous depth averaged vertical velocities of daily segments of ADCP measurements between 350 m and 50 m, following Jiang et al. (2007). Sunrise and sunset times are superimposed. (b) Average of the three highest upwards and downwards speed values per day. The hours of fast zooplankton motion are also shown.”.

Fig.9- The velocity unit on the reference coloured bar is missing. For each deployment, the data displayed are time-averaged, I suppose. This has to be clearly indicated.

Reply: The plots are histograms to visualise data density. The color bar show number of measurements inside each bin (the units were added next to the bar). The averaged velocities are also plotted with the thick lines. The legend was changed to clarify the figure. “The time average velocity at each depth is superimposed” was added to the caption.

Fig. 10- The events of harsh weather reported in section 3.4 should be indicated on panel e).

Reply: The events of harsh weather were shaded on all panels. “Grey shaded areas denote the three harsh weather events referred in the text.” was added to the caption.

Fig.11- It is nice but not necessary because it shows a qualitative snapshot (1 sample) of the zooplankton community captured with a 500 μm Bongo net, not useful enough to interpret the ADCP data.

Reply: The figure was removed from the manuscript. The corresponding text was modified in the discussion.

References The Mann&Lazier book is reported as 2005 edition here and 1991 in the text (pg.2, L1).

Reply: The text reference was changed to 2005.

REPLIES TO REVIEWER 2

We thank the reviewer for carefully reading the manuscript and providing useful comments.

The paper “ADCP observations of migration patterns of zooplankton in the Cretan Sea” by E. Potiris et al. presents the analysis of about 2.5 years of acoustic data from several deployment of an ADCP (RDI, 75 kHz) on a sub surface mooring in the Cretan Sea in order to infer the migration patterns of the zooplankton population in that area. The analysis is based on raw backscatter acoustic data from ADCP and ancillary data collected by a fixed open ocean observatory (E1-M3A), during cruises or from external data centers. An interesting aspect of the paper is the demonstration that ADCP data collected for other "standard" purposes (namely investigation of currents in the water column) can be also used to gather biological information (e.g., vertical migration of zooplankton). This might give new life to already existing dataset, not completely exploited yet. The paper is very well written, clear, easy to read and to understand.

The paper starts describing the processing method and the data, the environmental conditions (both during the experiment period and with a climatic perspective). The discussion about the backscattering data and the reasoning about the different groups of organisms is really amazing and nicely justified. Although, unfortunately, not fully supported by in-situ catchments, the authors were able to properly correlate acoustic data with biological information as well as with meteorological conditions. Just the latter analysis is one of the novel finding of the paper that is not usually in other biological- inspired works. The only significant migratory pattern evidenced is the normal one, with zooplankton species going toward deeper layers during the day and going upward toward the surface to feed during the night. The contribution of moon phases, as already evidenced by other cited studies, was also taken into account and discussed.

The distinction of the four groups of zooplankton (A,B,C,D) is based only on a visual analysis of Sv values; maybe it could be useful to analyze the vertical velocities at different depths to better identify the migratory pattern of each group.

Reply: Vertical velocities were analysed along with the backscatter coefficient at different depths. The analysis resulted in the update of some of our previous results. The updates were incorporated in the manuscript in the Results section.

5 SPECIFIC COMMENTS

Throughout the manuscript several symbols ">", "<", "~" are used. I would suggest to replace such symbols with words, i.e. greater, lower, about etc.

10 Reply: The mentioned symbols were replaced with words as suggested.

Title of section 2 and 2.2 have some capitalized letter. Please make them uniform with the other titles.

Reply: The titles were made uniform.

15

Page 5, line 10. The unit NM (nautical mile?) seems to refer to a speed. Likely it should be changed to knot.

Reply: The whole paragraph was removed as suggested by reviewer 1.

20

Page 5, table 2. It includes the pH parameter which is not mentioned in the text nor used in the description of the ancillary data. I think it is worth discarding it.

Reply: The pH parameter in the table was discarded as suggested.

25

Page 7, line 5. Correct the reference to Figure 3: "Figure 3Figure b&c".

Reply: The reference to Figure 3 is now "Figure 3, b & c".

30

Page 6, figure 2. Although the background color of the plots in the third row is grey, the used color scale doesn't allow to appreciate all lines. Especially those plotted in white are almost invisible. Please, consider to change the color scale.

Reply: The color scale was changed as suggested.

35

Page 8, figure 5. The labels above the plots and the caption seems not to agree with respect to the full moon and new moon.

Reply: The caption was corrected: "The yellow and black circles...".

40

Page 12, figure 8. Caption is not clear and, for example, it might be re-arranged as follows: (a) Instantaneous depth averaged vertical velocities of daily segment of ADCP measurements between 350 m and 50 m, following Jiang C2 et al. (2007). Sunrise and sunset times are superimposed. (b) Average of the three largest speed measurements per day.

45

Reply: Caption was re-arranged as "(a) Instantaneous depth averaged vertical velocities of daily segments of ADCP measurements between 350 m and 50 m, following Jiang et al. (2007). Sunrise and sunset times are superimposed. (b) Average of the three highest upwards and downwards speed values per day. The hours of faster zooplankton motion are also shown".

50

Page 13, figure 9. The unit of the color bar is missing.

Reply: Color bar units were added to the figure.

ADCP observations of migration patterns of zooplankton in the Cretan Sea

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Abstract. The lack of knowledge of the mesopelagic layer inhabitants, especially of those performing strong vertical migration, is an acknowledged challenge as its incomplete representation leads to the exclusion of an active carbon and nutrient pathway from the surface to the deeper layers and reversely. The vertical migration of mesopelagic inhabitants (macroplanktonic and micro-nektonic) was observed by acoustical means in the epi- and mesopelagic layer of the open oligotrophic Cretan Sea ([south Aegean Sea](#), Eastern Mediterranean) for almost 2.5 years at the site of an operational fixed-point observatory located at ~~1500m~~[1500 m](#) depth. The observed organisms were categorized in four groups according to their migration patterns. The variability of the migration patterns was inspected in relation to the physical and biological environmental conditions of the study area. The stratification of the water column does not act as a barrier for the vertical motion of the strongest migrants, moving up to 400 m every day. Instead, changes of light intensity (lunar cycle, daylight duration, cloudiness) and the presence of prey and predators seem to explain the observed daily, monthly and seasonal variability. The continuous presence of these organisms, yet capable of vertical motion and despite the profound seasonal circulation variability at the site of the observatory, implies their presence in the broader study area. The fundamental implications of the above for biogeochemical processing in oligotrophic seas due to the intimate link of the C and nutrient cycles, are discussed.

1 Introduction

The ~~inhabitants~~[biological organic carbon pump](#) is the major oceanic process that photosynthetically converts the dissolved CO₂ in the surface layers of the ~~deep sea~~[play an important role in ocean](#) to particulate organic carbon, which is then consumed by pelagic biota, and exported to depth by a combination of sinking particles, advection or vertical mixing of dissolved organic matter and transport by animals (Turner, 2015). Buesseler and Boyd (2009) have noted that “the surface ocean” is “where the ‘strength’ of the biological pump is set” whereas “the subsurface ocean” is “where the ‘efficiency’ of the biological pump is determined.”

The ~~main factor~~[determining the depths to which carbon is exported, a role mainly played by microbes and biological pump’s efficiency in the mesopelagic and bathypelagic zones](#), is the organic carbon export done by sinking and vertical migration by zooplankton and fish, together with microbial degradation. The diel vertical migration (DVM) of zooplankton, i.e. their vertical movement in the water column within the 24-h day, creates an active flux, pumping carbon between epipelagic and mesopelagic layers (review by Turner, 2015). ~~However, the lack of data from~~[This DVM of zooplankton has as its most common pattern a single descent to maximum depth during daytime, and a single ascent at minimum depth during night-time](#) (review by Cohen and Forward, 2009). DVM is a behavioural response that has been related to several exogenous (light, temperature, salinity, oxygen, hydrostatic pressure, stratification) and endogenous (sex, age, state of feeding, biological rhythm) factors (review by Forward, 1988; van Haren 2014, and references therein). Light has emerged as the major external factor controlling DVM behavior (review by Cohen and Forward, 2009), although insufficient to explain some cases where DVM occurs at depths larger than 1000 m, where light cannot penetrate (van Haren 2014, and references therein).

There are still gaps of knowledge regarding midwater depths which severely limits our ability to quantify the efficiency of the biological pump (Robinson et al., 2010). Moreover, the primary production at the surface ocean, which is the process that controls the strength of the biological pump, depends on nutrient cycles, and this dependence is stronger in oligotrophic seas. However, our knowledge of this process is limited in the open ocean, due to limited observations.

In the open Mediterranean Sea, and especially the oligotrophic Eastern Part, our understanding of the CO₂ functioning is essentially based on epipelagic observations or biogeochemical (BGC) model results. In fact, although there have been several studies on vertically migrating zooplankton in the Mediterranean Sea, little is known for its eastern part (review by Saiz et al., 2014). The scarce, short term, observations have reported different migration strategies of zooplankton (Fragopoulou and Lykakis, 1990; Koulouri et al., 2009) and dependence upon hydrology and food availability (Nowaczyk et al., 2011).

The open Cretan Sea's biochemistry is representative of a wide area of the Eastern Mediterranean ($0.6-1.6 \times 10^6 \text{ km}^2$ depending on the parameter) (Henson et al., 2016). In the Cretan Sea, the shifted seasonal cycle of primary productivity (Psarra et al., 2000) and the presence of a deep layer of high chlorophyll concentration, a common feature of the Mediterranean Sea, referred to as the Deep Chlorophyll Maximum (DCM hereafter) in the bibliography (Kimor et al., 1987; Yacobi et al., 1995), are examples of the close coupling of biological and physical processes in the broader study area. In fact, although the DCM's magnitude is mainly determined by biological mechanisms, the DCM depth and structure are essentially determined by optical-physical factors (e.g. Mann and Lazier, 1991; Concerning, Varela et al., 1994; Crise et al., 1999). The vertical extent and the intensity of the two mesoscale gyres forming a dipole in the Cretan Sea (Theocharis et al., 1999; Cardin et al., 2003; Kassis et al., 2015) govern to a large extent this coupling (Petihakis et al., 2002).

The range of zooplankton size from less than 2 μm (for heterotrophic nanoflagellates) to several meters (large jellyfish), makes the complete representation of all its components in a single net sample difficult to achieve (Frangoulis et al., 2016 and references therein). In the Mediterranean Sea, zooplankton DVM, one of the methodological limitations, is generally captured with a standard 200 μm mesh size net. However, that the populations of large individuals (macrozooplankton) belonging to important and common predatory groups, capable of strong migrations, such as the chaetognaths, amphipods, euphausiids, decapods, have been underestimated, as many of them escape the standard 200 μm mesh size net when towed at the recommend speed of $< 1 \text{ m s}^{-1}$ (Moriarty and O'Brien, 2013).

However, such large individuals can be detected by low frequency Acoustic Doppler Current Profilers (ADCPs hereafter). In fact, progress

Progress in ocean acoustics and marine technology during early 1980s (Holliday, 1977; Holliday and Pieper, 1980; Holliday et al., 1989; Costello et al., 1989) allowed the estimation of distribution patterns and biomass of zooplankton and micronekton to be inferred from ADCPs (Flagg and Smith, 1989). To measure currents, ADCPs transmit sound pulses in different directions. The sound is scattered by particulate matter in the water column, and radial velocities are computed from the Doppler shift of the backscattered signal (Gordon, 1996). The intensity of the backscattered sound can also be used in conjunction with net samples for estimating the biomass of zooplankton (Ashjian et al., 2002). Although the estimated ADCP backscatter is a by-product (Bozzano et al., 2014) and thus more suitable for qualitative than quantitative analysis (Brierley et al., 1998), field studies complemented with ADCP-derived sound scattering have been used to describe biological patterns in the interior of the ocean, such as zooplankton aggregations (Zhou and Dorland, 2004) and vertical migration (Postel et al., 2007) with remarkable detail.

In the Mediterranean Sea, there have been several studies on vertically migrating zooplankton (see table 5 in Andersen et al., 2001) and some have shown a substantial contribution to total carbon export in deep waters by the active transport of migrants (Isla et al., 2015, and references therein). However, these Mediterranean DVM studies have been mainly based on net sampling for short total sampling periods (e.g. discontinuous sampling at monthly frequency or continuous sampling for a few weeks). It is worth noticing that studies of macrozooplankton are less than of mesozooplankton, and that even for mesozooplankton there are very few interannual scale studies in the open Mediterranean Sea (Siokou-Frangou et al., 2010).

Among the Mediterranean DVM studies, the majority has been made in Western Mediterranean (see table 5 in Andersen et al., 2001; review by Saiz et al., 2014). These studies have pointed out different migrating strategies depending on species, with some species migrating upwards and other downwards at night, as well as a within species age-depth and sex-depth differential distribution. These observations were made in both the Eastern and Western Mediterranean (Fragopoulou and Lykakis, 1990; Andersen et al., 2001). The only DVM study in the Cretan Sea (Heraklion Bay) upper slope (300 m seabottom) studied near-bottom macrozooplankton, and reported a reversed DVM, i.e. downward migration during night (Koulouri et al., 2009).

The Cretan Sea is the most voluminous and deep (2500 m) basin of the Aegean Sea and an area of intense mesoscale variability and multiple scale circulation patterns. Two mesoscale gyres form a dipole in the Cretan Sea (Theocharis et al., 1999; Cardin et al., 2003; Kassis et al., 2015), while it is also an area of intermediate and/or deep-water formation (review by Skliris et al., 2014). Such areas of water formation are key locations for monitoring of the Mediterranean biochemical functioning (Malanotte-Rizzoli et al., 2014). Modelling studies have shown that the open Cretan Sea's biochemistry is representative of a wide area of the Eastern Mediterranean varying from $0.6\text{--}1.6 \times 10^6 \text{ km}^2$ depending on the variable (i.e. PP, SST, pH, O_2 , NO_3 , Chl- α) (Henson et al., 2016).

Satellite (SeaWiFS) studies of chlorophyll- α (Chl- α) have clustered the Cretan Sea in a wider bioprovince of the South-Central and Eastern Mediterranean that covers 60% of the total Mediterranean area, characterized by generally oligotrophic conditions (D'Ortenzio and Ribera d'Alcala, 2009). The presence of a Deep Chlorophyll Maximum (DCM hereafter), a common Mediterranean feature, which deepens going eastwards (e.g. Lavigne et al., 2015), also characterises the Cretan Sea and depends on a close coupling of biological and physical processes. In fact, although the DCM's magnitude is mainly determined by biological mechanisms, the DCM depth and structure are essentially determined by optical-physical factors (e.g. Mann and Lazier, 2006; Varela et al., 1994; Crise et al., 1999). In the Cretan Sea the vertical extent and the intensity of the mesoscale gyres forming a dipole govern to a large extent this coupling (Petihakis et al., 2002).

During Zooplankton studies in the Cretan Sea, were all done only on mesozooplankton (Mazzocchi et al., 1997; Siokou-Frangou et al., 1997; Siokou et al., 2013), with only one study on near-bottom macrozooplankton (Koulouri et al., 2009). Mesozooplankton abundance in the Cretan Sea's epipelagic layer was found to be at the same level as in the open Ionian and Levantine and community composition have shown significant similarities with the above areas (Siokou-Frangou et al., 2010 and references therein). Regarding deep living zooplankton, less is known for the Eastern Mediterranean (review by Saiz et al., 2014), although deep living mesozooplankton stock and composition has been investigated (Siokou et al., 1997, 2013) and its importance in consuming sinking particles reported (Koppelman et al., 2004). For deep living macrozooplankton there is even less information. The euphausiid species found in the whole Mediterranean Sea are the same, however the predominant ones are different in the eastern basin and Thyrrenian Sea, than those found in the western basin west of the Thyrrenian Sea (Wiebe and Abramo, 1972).

ADCP studies of DVM the Mediterranean have been limited. In the central Ligurian Sea, Bozzano et al. (2014) used an upward looking ADCP positioned at 100 m depth. They found that the main migration pattern is the daytime migration, and also, that deeper and stronger migration ranges are encountered during winter. In the Alboran Sea, van Haren (2014) with an upward looking ADCP positioned at approximately 800 m depth, found DVM to be related with internal waves. To our knowledge, there has been no ADCP study of DVM in the Eastern Mediterranean. However, during a study of the current velocities in the Cretan Sea back in 2000, Cardin et al. (2003) using the same ADCP (75 kHz), as in the present study, deployed in the same location, Cardin et al. (2003) reported a noise in the measurements of vertical velocity. In fact, at 75 kHz, and migrating zooplankton was given as a possible explanation.

The present study was stimulated by this hypothesis by Cardin et al., and knowing that for the 75 kHz frequency objects with size of 5 mm (1/4 of transmit pulse wavelength) or more, reflect sound and, thus cause causing a strong backscatter signal (Thomson and Emery, 2001). The migrating zooplankton was given as a possible explanation by Cardin et al. at the time.

During the present study ~~Following this hypothesis~~, four consecutive deployments of ~~at the same~~ 75 kHz ADCP ~~at the site of an operational open sea observatory in the Cretan Sea, and at the same location as Cardin et al.,~~ covering a period of two and a half years, provided a unique opportunity to study the migration patterns of zooplankton, continuously and in high frequency, for a long period in relation to environmental conditions. The aim of this paper is to present the observed distribution patterns of zooplankton (focusing on ~~diurnal migration~~), DVM) and discuss their relation to physical and biological environmental conditions, such as daylight, currents, stratification and food resources, ~~and to provide new information useful for zooplankton studies in the Cretan Sea, while opening a new way to evaluate an active pathway of carbon cycle of the wider area.~~

2 Materials and ~~Methods~~ methods

2.1 Experimental setup

The Poseidon E1-M3A observatory (www.poseidon.hcmr.gr) is located at 25.12° E, 35.74° N in the center of the Cretan Sea (~~south Aegean Sea~~) at a depth of 1500 m (Figure 1, a). It consists of two moorings: the first one has a surface buoy and a real-time multi-sensor array down to 1000 m, while the second is a subsurface one, hosting an upward looking RDI 75 kHz ADCP. The observing effort is complemented by monthly R/V cruises to perform CTD casts ~~and water plankton sampling (temperature, salinity, fluorescence) and net tows (zooplankton)~~. The ADCP data set used in the present study consist of four successive deployments of variable duration, which extended over a period of two years and ~~sevensix~~ months, from 15 November 2012 to 20 May 2015 (Table 1). The distance between the ADCP mooring line and E1-M3A buoy varied slightly but it was less than 2.7 NM for all deployments (Figure 1, c). The primary purpose for the ADCP deployment was to study currents. ~~The~~ The first deployment was considered as a test of the setup, so the sampling scheme and the depth of the ADCP were selected empirically (Table 1). The first deployment confirmed the feasibility of monitoring biological activity and it became obvious that a greater depth should be chosen, since the parking depth of zooplankton was deeper than the initial ADCP deployment depth and the sea surface reflection contaminated the first 50 m of the water column. However, the rearrangement of the mooring line was not possible due to the tight schedule of the next two deployments, and thus, only the last ADCP deployment was ~~~about~~ 120 m deeper than the previous ones.

The ADCP sampling plan was optimized in terms of temporal and spatial resolution by setting different sampling schemes at each deployment (Table 1; Figure 1, b). The aim was to check the consistency of the vertical velocity measurements of zooplankton and backscatter coefficient (defined in Sect. 2.2) between deployments. No significant differences in the vertical velocities and the backscatter coefficient between deployments of variable cell length and sampling rate, is an indication of reliable – accurate measurements. Thus, it is possible to identify biases caused by the sampling scheme, instead of the velocity errors due to the ADCP accuracy and of the backscatter coefficient estimation methodology.

One parameter used to ~~characterize zooplankton behavior as well as to~~ potentially identify the optimal sampling strategies ~~cell extension and sampling interval for the most appropriate recording of zooplankton signals~~ was the hereafter defined *burst velocity* ~~speed~~. The burst ~~velocity is~~ speeds of each cell are defined as the ~~maximum of the several 30-min or 1-hour~~ highest and lowest vertical velocity ~~average values~~ measurements respectively during ~~each ascent or each descent of the zooplankton population.~~

a time period of one day. The velocity ~~estimation~~ measurement inside a cell over the sampling interval is the result of the averaging of several pings. ~~Regarding~~ As the time resolution recording interval increases, the cell extension decreases and the actual zooplankton speed increases, we expect the actual zooplankton speed to be underestimated because zooplankton will not be inside the cell throughout the duration of the measurement but only during a fraction of it. The largest underestimation is expected when the actual zooplankton migrating speed is maximum. Thus, comparison of upward and downward burst

velocities between deployments at depths around 250 m (the depth at which the highest migrating speeds were recorded) were used to identify the most appropriate sampling scheme.

The challenge was to identify the lowest sampling rate that would still give acceptable resolution of the ascending/descending zooplankton movement, while conserving power and extending the deployment period as much as possible. Two sampling and averaging intervals, of lengths 30 min and 1 hour respectively, were tested in order to select the optimum sampling scheme. During the first deployment, a sampling interval of 30 min was used, to be followed by a 1 h interval during the second deployment. Comparison of the data from the two deployments revealed an underestimation of burst migrating velocities during the second data set (Figure 2, a & b) because of the lower sampling rate (1 h), thus the initial value of 30 min was selected for the last two deployments.

The range of the cells used (10 m – 20 m), on the other hand, did not affect the burst velocity and the average velocity measurements. Based on visual inspection, smaller cell extension during the 1st, 3rd and 4th deployments (30 min sampling interval) did not result in smaller burst and average speeds. However, using a small bin size (10 m) during the last deployment resulted in noisy velocity measurements. The depth-integrated S_v (backscatter coefficient, defined in Sect. 2.2) between the depths observed at all deployments were also consistent. The seasonal variability of the physical properties of the water column affected the estimation of S_v at depths shallower than 100 m, mostly during late August. Placing the ADCP at an upward looking position at a smaller depth than the nominal range resulted in erroneous data of the first 50 m of the water column due to sound reflection on the sea surface.

2.2 Data processing/analysis and visualization of ~~Backscatter~~ backscatter data

The backscatter coefficient S_v [dB re ($4\pi \text{ m}^{-1}$)] is given as:

$$S_v = C + 10\log_{10}((T_x + 273.16)R^2) - L_{DBM} - P_{DBW} + 2aR + K_c(E - R_r), \quad (1)$$

where C [dB] = -159.1 is an instrument constant, T_x [°C] the transducer temperature, R [m] the slant range, L_{DBM} the $10\log_{10}$ of the transmit pulse length [m], P_{DBW} the $10\log_{10}$ of the transmit power [W], a [dB m⁻¹] the sound absorption coefficient, K_c a constant of proportionality for converting the incoming raw echo data to dB, E [counts] the raw echo data and $E_r = \min(E)$ [counts] the reference raw echo per transducer when there is no signal. S_v was calculated according to Deines (1999), K_c according to Heywood (1996), the speed of sound for the calculation of R according to Gordon (1996) and a according to Ainslie and McColm (1998).

Instantaneous vertical velocity profiles were depth averaged and split in daily pieces/datasets to identify the hours of the day during which the zooplankton moves upward or downward, as well as the long-term seasonal and interannual variability in the ascend/descend hours. Another step was to select the maximum and minimum vertical velocities of each daily piece of data during the time of the upward and downward movement. Depending on the sampling rate, two to four samples were averaged. At last, histograms of vertical velocity versus depth one hour before and one hour after sunrise/sunset times were used to identify possible ascending/descending differences of migration patterns and evaluate the consistency between the different ADCP sampling schemes.

Climate Data Operators (CDO, 2018), Ocean Data View (Schlitzer, 2016) and Generic Mapping Tools (Wessel et al., 2013) were used for the data processing and visualization. Wind stress and sensible/latent fluxes were computed from the quality-controlled buoy data with the air-sea toolbox to identify the time when conditions favor overturning of the water column (http://woodshole.er.usgs.gov/operations/sea-mat/air_sea-html/index.html).

2.3 Description and processing of auxiliary data

To estimate volume backscattering, assess environmental conditions during deployments and assist interpretation of ADCP measurements, several complementary data sets have been used (Table 2).

The E1-M3A buoy measures meteorological variables (wind speed, gust and air temperature were used here) as well as temperature, conductivity and fluorescence at multiple depths (20, 50, 75 and 100 m). Temperature and conductivity measurements are also available at the sea surface and at 250 m depth. The chlorophyll concentrations are measured with the WETLABS ECO FLNTU fluorescence sensors which are mounted on the moored 16plus CTDs. A downward looking Nortek 400 kHz ADCP is mounted to the buoy hull, measuring horizontal currents at 5 m bins from the surface down to 50 m depth. Due to the lack of compatibility of the 400 kHz ADCP with the buoy software, backscatter measurements are not available from this instrument. The above meteorological and marine surface parameters were downloaded from the Poseidon on-line database (www.poseidon.hcmr.gr), where they are stored in real-time. In addition, due to occasional problems with the real-time underwater transmission, subsurface sensor data were downloaded from the memory logs of the instruments during the regular bi-annual maintenance. Meteorological and sea surface measurements from the buoy's sensors span a period of 24 Mmonths (from 22 May 2013 to 25 May 2015) and subsurface measurements a period of 20 Mmonths (from 22 May 2013 to 10 January 2015). Buoy data have undergone automated quality control, such as rejection of stalled values and application of min-max and spike filters. Visual inspection was the last quality control step; the remaining suspect measurements were removed manually. The heat flux through the air-sea interface computations were based on the air-sea interaction Matlab routines provided by Rich Pawlowitz (via the SEAMAT collection, <https://sea-mat.github.io/sea-mat/>), applied on the E1-M3A meteorological and sea-surface data.

2.4 Limitations

Several limitations of the ADCP and auxiliary data should be carefully considered. S_v is a proxy for zooplankton biomass and when integrated along the acoustical beams it can provide a gross measure of the instantaneous biomass of the water column (changes in the acoustical character of zooplankton cannot be identified). Although the integrated S_v is consistent among the deployments (discrepancies between deployments were observed only for the first few bins), such an analysis is not meaningful with the experimental configuration of this study, since the zooplankton is not permanently in the range of the ADCP and because of the seasonal succession of dominant species constituting the zooplankton stocks in the Cretan Sea (Gotsis-Skretas et al., 1999). The upper 50 m of the water column are not measured, thus the depth integrated S_v exhibits significant variability due to the monthly change of the depth at which zooplankton ascends during night-time because of moonlight. Also, the whole deep scattering layer is found inside the ADCP range only for a small period of the 4th deployment, adding another source of variability that is not attributed to biomass changes of zooplankton. Another source of error, which largely depends on the availability of auxiliary data, is the imperfect calculation of the effects of the gradients of the upper water column in the estimation of S_v due to the changes in temperature and salinity. The above problems are generally encountered when measuring zooplankton with upward looking ADCPs and should be treated with caution in acoustical studies of zooplankton.

~~At the E1-M3A zooplankton samples are taken regularly (monthly) with parallel tows of two nets (45 μ m mesh size and 200 μ m mesh size) from 100 m to the surface. To validate the ADCP measurements a field sampling strategy targeting zooplankton organisms at the size of 5 mm would require day and night multilayer net tows down to 500 m depth, with mesh size >200 μ m towed at a speed >2 NM. Unfortunately, logistically this proved not feasible and only on 19/12/2013 at 12H00 (local time) one oblique tow at a speed of 2 knots with a Bongo net equipped with a 330 μ m and a 500 μ m net (both with a flowmeter) was performed. As this was possible only once, the information obtained is considered as an indication of the type of large~~

metazoans present above the ADCP at the sampling time, but cannot be considered representative of the average relative abundance of migrants in the area and thus mentioned briefly in the discussion.

3 Results

3.1 Environmental conditions at the study site

2.5 Hydrology of the Cretan Sea

The ADCP site is located at the center of the semi-permanent dipole of the Cretan ~~sea~~Sea, which consists of a cyclone to the east and an anticyclone to the west of the observatory (Theocharis et al., 1999; Korres et al., ~~2014~~2014). Low frequency variability at the study site is controlled by the intensity and the vertical extent of the dipole as reported by Cardin et al. (2003). Four water masses fill the surface and subsurface layers of the Cretan ~~sea~~Sea. Modified Atlantic Water (MAW, $S=38.5\text{--}38.9$ psu) fills the 20 m- ~~100~~ 100 m layer. Cretan Intermediate Water and Levantine Intermediate Water, that have similar characteristics (CIW & LIW, $\theta=14.9\text{--}15.1$ °C, $S\sim 39\text{--}39.1$ psu) fill the 200 m- ~~500~~ 500 m layer. Transitional Mediterranean Water (TMW, $\theta=14.2$ °C, $S=38.92$ psu), a mixture of Levantine Intermediate Water and Eastern Mediterranean Deep Water enters through the Cretan Straits and its core lies at the 500 m- ~~800~~ 800 m layer (Georgopoulos et al., 2000; Velaoras et al., 2013) or deeper (Velaoras et al., 2015). Below TMW lies the Cretan Deep Water, a water-mass argued to have local (Theocharis et al., 1999) or North/Central Aegean origin (Zervakis et al., 2000; Gertman et al., 2006). Inflow of Atlantic water (Theocharis et al., 1999), typically during late summer, causes ~~The temperature at the depth of the deep scattering layer ~450 m (based on the available data set; Figure 2, d, e & f), ranges from 14.55 °C to 14.9 °C and the salinity from 38.98 psu to 39.04 psu~~ salinity minimum at the subsurface layer.

3 Results

3.1 Environmental conditions at the study site

Sea surface temperature ~~ranges~~ ranged seasonally from 15 °C to 26 °C and salinity ~~ranges~~ ranged from 38.8 psu to 39.5 psu (Figure ~~23~~ 23, b, c). The salinity of the deeper layers ~~ranges~~ ranged from 38.9 psu to 39.1 psu. Lowest temperatures ~~are~~ were observed during February and March, while highest temperatures during August and September. The seasonal cycle of temperature ~~penetrates~~ penetrated down to 100 m and the permanent thermocline ~~extends~~ extended down to 350 m (Figure ~~23~~ 23, b). Salinity also ~~exhibits~~ exhibited a seasonal cycle down to 100 m but the seasonal signal ~~dominates~~ dominated salinity variations of the upper part of the water column (Figure ~~23~~ 23, c). Largest salinity values ~~are~~ were observed during calm, cloud free summer days, ~~due to intense evaporation and the intrusion of high salinity surface water of Levantine origin (Theocharis et al., 1999).~~ A ~~Inflow of Atlantic water (Theocharis et al., 1999), typically during late summer, causes~~ the salinity minimum at the subsurface layer from 40 m to ~~between surface and~~ 100 m ~~depth was also observed~~ in Figure ~~23~~ 23, c-~~& e~~ e. Deep casts (Figure ~~23~~ 23, e & f) ~~reveal~~ revealed a continuous change of the water column towards fresher and colder values between 250 and 1000 m especially from 2012 to 2016, which points to intensified horizontal motion of the subsurface layers. The temperature at the depth of the deep scattering layer around 450 m (based on the available data set; Figure 3, d, e & f), ranged from 14.55 °C to 14.9 °C and the salinity from 38.98 psu to 39.04 psu.

At the study area, prevailing winds ~~blow~~ blew from N-NW (Figure ~~34~~ 34, a). Short-term variability of air temperature during winter ~~is~~ was larger, due to strong northerly winds, which ~~cause~~ caused the air temperature to drop below 10 °C. Latent heat loss typically ~~ranges~~ ranged between 100 ~~W m⁻²~~ W m⁻² and 200 ~~W m⁻²~~ W m⁻². Sensible heat flux also ~~results~~ resulted in net loss, but

it ~~iswas~~ negligible from March to October. During the rest of the year typical values ~~arewere~~ less than 50 WmW m^{-2} . Sensible heat loss ~~contributescontributed~~ significantly to the flux budget when wind stress ~~becomesbecame~~ larger than 0.2 Pa (~~typical values are average wind stress of the complete data set was~~ 0.482 Pa). Wind stress during December of 2013 was more than 0.2 Pa on average, while peak values of 0.8 Pa were also observed. Consequently, the monthly average sensible heat flux was about 100 WmW m^{-2} and peak values were about 300 WmW m^{-2} . Latent heat during that period peaked at 600 WmW m^{-2} . Similar atmospheric conditions that ~~favorfavoured~~ convection of the upper 100 m of the water column were observed during 10 to 22 February 2015, ~~a period during which changes in the vertical distribution of zooplankton were observed.~~

Average water velocity from surface down to 50 m ~~iswas~~ 0.29 msm s^{-1} towards S-SE and is invariant with depth. The layer between 50 m and 350 m ~~isdepth was~~ characterized by a diminishing vertical shear that ~~iswas~~ largest between 50 m to 150 m ~~depth~~ and ~~vanishesvanished~~ below 400 m (Figure 34, d, where only the fourth deployment is displayed, since in previous deployments the larger bin size caused underestimation of the high vertical wavenumber shear). The average current speed below ~~the depth of~~ 350 m ~~iswas~~ 0.06 msm s^{-1} . The direction of the axis of maximum variance between the surface and 50 m ~~iswas~~ S-SE and gradually ~~turnturned~~ to S-SW at 200 m ~~depth~~. Currents ~~arewere~~ less unidirectional with depth too. The strong currents of the surface layer ~~exhibitexhibited~~ the least directional variability (Figure 34, b & c). High frequency variability at the site ~~consistsconsisted~~ of inertial and tidal currents, which ~~accountaccounted~~ for a small portion of the total variance (less than 8-%), even though the inertial motions ~~arewere~~ dominant over short periods. ~~Low frequency variability is controlled by the intensity and the vertical extent of the dipole as Cardin et al. (2003) have mentioned.~~

During the study period the core of DCM was observed between 70 m and 120 m and its vertical extent was ~~around~~ 60 m (Figure 45, a). On average, the largest chlorophyll values ~~arewere~~ observed at the 75 m and 100 m sensors of the buoy (Figure 45, b). Also, at these depths, the short-term variability ~~iswas~~ comparable to the variability due to the annual cycle, while for the 20 m and 50 m sensors the seasonal variability ~~iswas~~ dominant. The DCM ~~starts to formformed~~ from February to April and ~~iswas~~ usually destroyed by October (see changes of the depth range for which chlorophyll concentration is above 70-% of the maximum value in Figure 45, a, ~~as well as Figure 4, b~~).

3.2 Scattering and migrating groups of organisms

The results of the four ~~ADCP~~ deployments of a total duration of 2.5 years (Figure 5; ~~Figure 6~~) ~~provideprovided~~ a wealth of information about the scattering organisms and their movement in the water column. Characteristics that are easily visible ~~in Figure 6~~ include the ~~constant~~ presence of a deep layer of scatterers (unfortunately visible only in the last deployment, since only then was the ADCP placed deep enough to record it), a ~~diel, a~~ seasonal and a monthly (moon) cycle.

A closer examination of daily backscatter patterns (Figure 6; ~~allows7~~), ~~allowed~~ the categorization of the scattering organisms in four groups according to their migration patterns, on the basis of distinguishable trails of volume backscatter measurements of the ADCP. Three of them ~~exhibitexhibited~~ a daily ~~migrationalmigrating~~ pattern, while the fourth ~~remainsremained~~ at a constant depth. The first group (group A hereafter) ~~doesdid~~ not migrate. It ~~iswas~~ found at 400 m - 450 m, and it ~~formsformed~~ a ~~permanent~~ deep scattering layer (Figure 5 ~~last deployment, Figure 6~~). ~~The width of this scattering layer varies, signifying that it does not consist only of scatterers of group A, but there are also migrating organisms, which have their parking depth there, and spend part of their time in other depths.7, c).~~

Group B (Figure 6) ~~follows7~~) ~~followed~~ the normal ~~daily vertical migrationDVM~~ pattern i.e. it ~~movesmoved~~ close to the surface at dusk, ~~feeds there during night~~ and ~~returnsreturned~~ to the parking depth at dawn where it ~~staysstayed~~ during the day. This group ~~spendsspent~~ the day-time at 400 - ~~m~~ - 450 m ~~depth~~ and the night-time between 150 m and surface (Figure 5, ~~last deployment-7~~). When at the bottom of the seasonally varying ~~feedingday-time parking~~ layer (60 - 160 m) ~~(Figure 6)~~, its vertical velocity ~~decreases, probably as a result of feeding activitydecreased~~, while still moving towards the surface. The

bottom of the ~~feedingday-time parking~~ layer ~~iswas~~ identified by the deceleration of upward movement and subsequent increase of S_v (Figure 67), as the zooplankton ~~spendsspent~~ more time in a particular cell when moving at smaller speed. The change of depth of the bottom of the ~~feedingday-time parking~~ layer of group B ~~iswas~~ in good agreement with the time variation of the depth of maximum chlorophyll concentration (~~Figure 4~~).

5 The backscatter coefficient at any certain depth, as long group B ~~iswas~~ above that depth, ~~iswas~~ larger during night-time compared to day-time (Figure 67). The exception to this rule ~~iswas~~ the deep scattering layer. The result ~~iswas~~ the “curtain” shape of S_v seen in Figure 67, which implies that a part of zooplankton that forms group B ~~spreadsspread~~ in the entire 050 m - 400 m water column while migrating. The smallest S_v values, close to the system noise floor, ~~arewere~~ observed between 250 - 300 m, when group B ~~iswas~~ found at the parking depth.

10 Between the depth of ~~200300~~ m and ~~250350~~ m, S_v never ~~fallsfell~~ close to the noise floor; (~~Figure 7, c~~), even in the absence of group B during day-time, pointing to the presence of scatterers and thus to a third group of organisms (group C hereafter). Group C ~~also migratesmigrated~~ from 350 m to 300 m or from 250 m to 200 m depending on the period of the year; ~~probably as result of the change in day time duration~~.

15 ~~InAt~~ shallower depths a fourth group ~~can be was~~ observed (Figure 67, Group D), which ~~spendsspent~~ most of the day-time in a depth of 180 m - 240 m and during the night it ~~movesmoved~~ to more shallow depths of 60 m - 90 m, where its trails ~~meetsmet~~ with those of group B. This ~~iswas~~ close to the depth where the layer with the largest concentration of phytoplankton throughout most of the year (~~Figure 4, 5~~) ~~iswas~~ observed. Its backscatter signal ~~iswas~~ not as strong as that of group B. Actually, its trail ~~iswas~~ easily distinguishable mostly during the time of its upward motion, as a secondary thin strong S_v trail, shallower than the one caused by group B, and less during the downward motion (Figure 67). This signal ~~iswas~~ present at all deployments, ~~but not throughout each deployment~~, and its characteristics, such as depth and slope, ~~arewere~~ consistent between deployments. ~~With close examination, we can exclude the possibility of a false/mirroring echo coming from group B, as the changes of the trails of group B are not reflected in the trails of group D. Moreover, the profiles of vertical velocities are distorted towards lower speeds at the depth where group D is observed, from 180 m to 240 m (Figure 7, a & b; Figure 9, all panels), while profiles of the time average S_v between 180 m and 240 m is also larger.~~

25 ~~An interesting outcome is that the depth of the feeding layer is also significantly affected by the phase of the moon. Full moon is accompanied by the deepening of the bottom of the feeding layer by more than 50 m as can be seen in the backscatter coefficient (Figure 5, mainly in months October to February) and vertical velocity graphs (Figure 7, a & b).~~

~~An interesting observation derives from the combination of variability of the bottom of the feeding layer which is mainly on monthly basis, with the variability of the parking depth which is only on seasonal basis. This was evident at least for group B which migrates a larger distance. In fact, the parking depth of group B changes seasonally following the variation of the feeding layer, however, it does not present a monthly variation (Figure 5). This implies that the distance that group B travels changes more on a monthly basis than on a seasonal basis. This is consistent with the monthly variation of the migrating velocities seen in Figure 7.~~

30 ~~The distinction of group B from group A appears from the daily variation of the thickness of the deep scattering layer (Figure 10, e). It becomes thinner and the backscatter coefficient at that depth becomes larger during day time compared to night time. We infer that group A (although remaining on average at the same depth) spreads during the night, taking advantage of the absence of group B and aggregates again during day. Thus, during the short time period that group B leaves the parking depth the whole deep scattering layer spreads. Then, after the two groups are separated and for as long as group A is separate from group B, the deep scattering layer progressively becomes thicker. As soon as group B returns to the parking depth, the deep~~

40 ~~scattering layer, now consisting of both groups, shrinks, becomes denser and thus its backscatter coefficient increases. This would be an expected behavior if group B preys on the migrating zooplankton that forms group A. Although it is not possible to synthesize a clear picture from the available data, the persistent observations that the deep scattering layer becomes thicker by 30 m to 50 m during night supports the above hypothesis. Although the deep scattering layer was only partly in the range~~

of the ADCP during most of the period of the 4th deployment, during March and April 2015 the scattering layer was shallow enough to be properly measured by the ADCP (Figure 5; Figure 10, e). A contiguous vertical motion of the deep scattering layer in addition to the diurnal spreading was observed. The extent of the vertical motion was ~100 m during March and ~50 m during April 2015 (Figure 10, e).

3.3 Migration timing, duration and velocity

Concerning the three migrating groups (B, C, D) there are no differences in the duration between fast upward and downward movement. The duration of strong migration did not change with time, with two hours spent each way (four hours in total) (Figure 8, a). The duration of migration is roughly the same for all migrating groups (B, C and D), as they start to move upwards at the same time (with B, D trails meeting at the feeding layer), although it must be mentioned that only four measurements are available during each upward/downward motion. The upward motion of group B begins at its parking depth of ~350 m and ends at the bottom of the feeding layer (Figure 7, a; Figure 9, b, d, f & h). The depth of maximum negative acceleration, which marks the beginning of downward motion (the velocities of groups B, C and D cannot be distinguished), is generally found shallower than the bottom of the feeding layer (Figure 9, a, c, e & g), suggesting that zooplankton moves shallower as it feeds. Descend ~~is~~ was symmetrical with respect to the sunrise. It ~~starts~~ started one hour before and ~~ends~~ ended one hour after the sunrise (Figure 8, a). Ascend ~~starts~~ started half an hour before and ~~ends~~ ended one and a half hour after the sunset (Figure 8, b). As mentioned, it is not possible to distinguish the velocity of the migrating groups B and D, since the velocity measured by the ADCP above 250 m is the result of the motion of both groups. Depth-averaged velocities during strong migration time ~~are~~ were about 3 cm s^{-1} (Figure 9). While the duration of the strong migration was constant, the migrating velocity changed seasonally, following the duration of day which, at 35° longitude, lasts 9.8 h on winter solstice and 14.5 h on summer solstice, as is clearly shown in Figure 8, b, despite the fact that they are quite noisy. Downward velocity was slightly larger than the upward velocity by almost 1 cm s^{-1} on average (Figure 8, b). Monthly variability was observed at the depths at which strong downwards migration started (speeds higher than 2.5 cm s^{-1}) (Figure 9). Also, in Figures 9, a).

Group B ~~does~~ and it can be seen that zooplankton ~~did~~ not migrate at a constant velocity (Figure 7; Figure 9). Largest with depth. Highest upward velocities, ~~close to~~ about 6 cm s^{-1} ~~are~~ were recorded between 200 m and 300 m. Largest Highest downward velocities ~~are~~ were recorded between 250 m and 350 m (Figure 9, 10). Since Group B ~~travel~~ travelled the largest distance in the course of a day ~~and goes deeper than group D~~, the largest vertical velocities recorded, especially between 200 m and 350 m, ~~must~~ might be due to the migration of group B. At the depth of ~200 m the ADCP ~~records~~ recorded relatively small vertical velocities, about 2 cm s^{-1} (Figure 7, a & b; Figure 9 & 10, all panels), which ~~distort~~ distorted the vertical profile that would be expected by group B, unless group B ~~decelerates~~ decelerated at the bottom of the photic zone. The dispersion of the vertical velocity around the average value at that depth ~~is~~ was much less than all the other depths. ~~While the cause is clearly biological, to date it was~~ (Figure 10, all panels).

It was not possible to ~~understand~~ distinguish the velocity of group B from the velocity of group C, as their S_v trails overlapped during migration. However, utilizing the S_v trails attributed to groups B and D it was possible to distinguish the velocity of group D. The time average vertical velocities and S_v relative to sunrise and sunset hours showed that secondary peaks of S_v attributed to group D were accompanied by a small increase in vertical speed (Figure 11, all panels). According to the time average velocity measurements in Figure 10, a & b, group D migrated at an average velocity of about 0.2 cm s^{-1} . The migrating velocity of group D calculated indirectly using the trails of S_v (Figure 11, c & d) was about 0.4 cm s^{-1} . The depth averaged migrating velocity of 3 cm s^{-1} recorded by the ADCP and attributed to group B was consistent with the indirect calculation of the migrating speed of this group based on the distance travelled and explain it using the available data, though

the spreading of group B and the presence of group C that stays around that depth could explain these observations to some extent: the duration of its migration (about 3.5 cm s^{-1}).

At 35° longitude, day time lasts 9.8 h on winter solstice and 14.5 h on summer solstice. So, the time interval that the zooplankton has at its disposal in order to feed varies significantly in the course of a year (Figure 8, a). This affects the velocity of group B, which changes seasonally as is clearly shown in the burst velocities of Figure 8, b, despite the fact that they are quite noisy. The phase of the seasonal cycles of burst speeds and day /night time almost coincide. Downward velocity is slightly larger than the upward velocity by $\sim 1 \text{ cm s}^{-1}$ on average (Figure 8, b). The largest burst speeds are recorded during spring, when the seasonal pycnocline starts to form. This is the period of the year that the phytoplankton (chlorophyll α) is spread quite homogeneously throughout the upper 160 m of the water column and the DCM is not formed yet (Figure 6). Thus, the seasonal changes of velocity are driven by the combination of short night time and low food resources found at a relatively small depth, the duration of night time being the governing factor.

3.4 Effect of an extreme meteorological event

Three successive events of harsh weather conditions were observed from 10 to 13, from 17 to 21 and from 23 to 25 February 2015. The sky was mostly overcast (Figure 4012, a), air temperature dropped at 7.5°C (Figure 4012, b), wind speed reached 15 m s^{-1} and wind gust exceeded 20 m s^{-1} (Figure 4012, c). The third event was shorter than the first two and caused an increase in air temperature. The homogenization of the water column prior to the first event did not exceed the 50 m (as shown from the E1-M3A time series) while the nearest in time available CTD cast on the 3rd of March revealed that the first 100 m of the water column were then homogenized. The zooplankton was distributed from the surface down to 350 m all day long (Figure 4012, e), especially during the first two events, although S_v remained larger during night-time compared to day-time above the depth of 300 m. Also, only small migrating velocities were measured especially during the first two events (Figure 4012, d). During the second event, the core of the deep scattering layer moved shallower at the depth of 350 m. After the third event, the pattern of the backscatter coefficient above 300 m returned back to the “normal” conditions, but the deep scattering layer remained generally shallower and moved coherently in the vertical direction from 450 m to 350 m until the 2nd of April (Figure 4012, e). Thus, the combination of low light level due to clouds and convective currents, triggered changes at the deep scattering layer were observed, which is found well below the maximum depth at which the overturning took place.

4 Discussion

4.1 Factors affecting zooplankton migration

DVM of zooplankton has been related to several exogenous and/or endogenous factors (review by Ringelberg, 2010). In the present study, light intensity seemed to be the major factor affecting the zooplankton migration on daily, monthly and annual time scales. Light can act as an endogenous (entraining circadian rhythms) or exogenous factor controlling DVM. Several hypotheses attempt to explain the exogenous role of light. According to the rate-of-change hypothesis, the variation in relative rate and direction of change in light intensity is the cue to initiate DVM, whereas light acts also for orienting and controlling DVM (Cohen and Forward 2009).

A scenario that has been proposed to explain DVM is that of a photobehaviour formed in order to avoid the damaging effect of solar ultraviolet (UV) radiation. The UV photoreceptors found on zooplankton have supported this (Williamson et al., 2011). However, this mechanism fails to explain the maximum depth of DVM in our case, since UV radiation in the eastern Mediterranean reaches its maximum value at a depth of 50 m (Tedetti and Sempéré, 2006).

Another approach to explain DVM proposes a photobehaviour attempting to balance the need of feeding, while avoiding visual predators. Therefore, DVM as a photobehaviour should consider the rate of change of light combined with the rate of change of food abundance and kairomones (released by predators and detected by zooplankton). In order to maximize the detection of downwelling light, DVM organisms have adapted their maximum visual sensitivities to wavelengths of about 450 - 470 nm (although species with photosensitivity in wavelengths larger than 470 nm sensitivity have been reported as an additional adaptation to bioluminescent emissions) (review by Cohen and Forward 2009).

According to the results presented here, during full moon the zooplankton prey almost 50 m deeper than during the new moon, a possible behavioral response to increased light conditions. Twilight effects on DVM using data from a downward looking 300 kHz ADCP measuring from the surface down to 80 m were also reported by Bozzano et al. (2014) in the Ligurian Sea.

Changes in migration depth and speed were also correlated to cloudiness. Amplitude changes of the extent of DVM due to changes in cloudiness can also be found in the results of Pinot and Jansá (2001). Cloudiness may have an indirect effect on migrants, since the phytoplankton production, and thus the available prey concentration, becomes lower under lower light conditions. In addition, the prey is spread downward due to convection. Thus, the migrants have to spread in a larger water column in order to obtain a sufficient amount of prey.

4 Discussion

Migration patterns of zooplankton have been observed by an acoustical method using a long term time series for the first time in the eastern Mediterranean. The presence of organisms >5 mm, that backscatter the signal of a 75 kHz ADCP was recorded from the surface down to ~450 m. The persistent observations of these organisms throughout the duration of all deployments at the same location, even though the circulation in the study area is quite variable (e.g. moving semi permanent cyclone-anticyclone dipole, Korres et al., 2014), suggests the widespread presence of different groups of organisms >5 mm in the Cretan Sea. These organisms were categorized in four groups according to their migration behavior, with three of the groups exhibiting vertical migration. A non migrating group (group A) that shares the 450 m horizon with two migrating groups (B and C) was found. A third migrating group (group D) parked at an average depth of 180 m. Similar results were obtained in the Arabian Sea using a 153 kHz ADCP by Luo et al. (2000) who observed two groups migrating simultaneously. The vertical distance between the migrating groups observed by Luo et al. (2000) was larger during the upward motion and smaller during the downward motion, which might explain why the 75 kHz ADCP used in this study could not distinguish the two of the migrating groups (groups B and D) during the downward motion. Average migrating velocities of $\sim 3 \text{ cm s}^{-1}$ and burst velocities of $\sim 6 \text{ cm s}^{-1}$ were measured corresponding to the first migrating group (B) and agree with the average and burst speeds reported in Ott (2005) and references therein. Migrating velocities of groups C and D appeared to be smaller than those of group B. Interestingly, the migrating velocities obtained were the same, whether the velocity was directly measured by the ADCP, or via the indirect calculation using the backscatter signal (350 m migration over 2 hours, i.e. $\sim 5 \text{ cm s}^{-1}$).

Daily dispersion, aggregation and small vertical motion of the deep scattering layer were observed. The examination of zooplankton aggregation is out of the scope of this study due to the necessary compromise between sampling rate and total deployment duration, which result in coarse data for this purpose. However, the diel vertical migration and the avoidance of predation are well known biological drivers of the intensification of patchiness (Folt and Burns, 1999). The spreading out of the non migrating group (A) at night during the absence of the migrating group (B), and its shrinking back during day time during the presence of B, suggests that these two groups are coupled in a behavioral relation.

Diel vertical migration of zooplankton has been related to several exogenous and/or endogenous factors (review by Ringelberg, 2010). In the present study light intensity was found to be the major factor affecting the zooplankton migration. Seasonal and monthly variability, evident in the backscatter coefficient and vertical velocity, was dictated by the duration of day time and

moon phase respectively. Twilight effects on migration patterns using data from a downward looking 300 kHz ADCP measuring from the surface down to 80 m were also reported by Bozzano et al. (2014) in the Ligurian Sea, while smaller amplitude changes of the extent of vertical migration due to changes in cloudiness can also be found in the results of Pinot and Jansá (2001). According to the results presented here, during full moon the zooplankton preys almost 50 m deeper than during the new moon, a possible behavioral response to increased light conditions.

Another factor affecting migration was prey (in terms of Chl- α) concentration and location. The seasonally varying zooplankton feeding layer extended from the surface down to a maximum depth of 160 m. The bottom of the feeding-day-time parking layer was found at an average depth of 100 m. It was recorded deeper from May to July and shallower from November to January. The upward motion of the migrating groups decelerated at the depth of the largest chlorophyll concentration. The observations suggest that the vertical gradients of temperature, salinity, density and horizontal currents affect the migration of zooplankton less than their feeding behavior. The largest vertical velocities were recorded during spring, when the seasonal pycnocline started to form. This was the period of the year that the phytoplankton (Chl- α) was spread quite homogeneously throughout the upper 160 m of the water column and the DCM was not formed yet.

The fact that the parking depth of the migrating zooplankton groups B and C (which is also the parking depth of the non-migrating group too) is found so deep (450 m), cannot be explained by light, phytoplankton prey concentration (since these are zero below 200 m), nor by a temperature, salinity or density gradient at that depth. Considering that at the parking depth of these groups the vertical shear practically vanishes, and the horizontal currents are the weakest ones recorded, it might indicate an active behavioral adaptation to minimize energy loss by maintaining their position at a depth with minimum turbulence.

~~The combined effect of wind, cloudiness and convection in the upper layer affected the migrating groups by reducing their vertical velocities. They spread in almost the entire water column, and did not migrate as deep as usual during day-time. While convective events were not observed down to 450 m, the deep scattering layer was uplifted by several tenths of meters and its daily vertical motion became larger. Shoaling of the deep scattering layer was observed during winter and deepening during summer. The overall picture is that the environmental conditions affect the migrating groups and the changes propagate to the non-migrating group.~~

~~Several limitations of the ADCP and auxiliary data should be carefully considered. S_p is a proxy for zooplankton biomass and when integrated along the acoustical beams it can provide a gross measure of the instantaneous biomass of the water column (changes in the acoustical character of zooplankton cannot be identified). Although the integrated S_p is consistent among the deployments (discrepancies between deployments were observed only for the first few bins), such an analysis is not meaningful with the experimental configuration of this study, since the zooplankton is not permanently in the range of the ADCP and because of the seasonal succession of dominant species constituting the zooplankton stocks in the Cretan Sea (Gotsis Skretas et al., 1999). The upper 50 m of the water column are not measured, thus the depth integrated S_p exhibits significant variability due to the monthly change of the depth of the feeding layer because of moonlight. Also, the whole deep scattering layer is found inside the ADCP range only for a small period of the 4th deployment, adding another source of variability that is not attributed to biomass changes of zooplankton. Another source of error, which largely depends on the availability of auxiliary data, is the imperfect calculation of the effects of the gradients of the upper water column in the estimation of S_p due to the changes in temperature and salinity. The above problems are generally encountered when measuring zooplankton with upward looking ADCPs and should be treated with caution in acoustical studies of zooplankton.~~

Another limitation is due to the insufficient in situ data from large zooplankton in the area.

4.2 Zooplankton sampling considerations

Local literature does not allow us to clearly identify the taxonomic composition of the migrating ~~populations~~assemblages found in the present study. The few published studies that have sampled zooplankton in the epipelagic and mesopelagic layer of Cretan Sea, were all done with vertical hauls ($\approx 1 \text{ m s}^{-1}$) of 200 μm mesh size nets (Mazzocchi et al., 1997; Siokou-Frangou et al., 1997; Siokou et al., 2013) and are ongoing similarly during the monthly monitoring program at the E1-M3A observatory. Thus, they are inappropriate to capture organisms at the size of 5 mm, which are the smaller organisms expected to contribute significantly to the backscatter of a 75 kHz ADCP. It is however clear that the ~~populations~~assemblages examined in our study include organisms other than copepods, since the biggest copepods species reported in the area (Mazzocchi et al., 1997; Siokou-Frangou et al., 1997; Siokou et al., 2013) reach a maximum size of $\approx 3.5 \text{ mm}$ (Razouls et al., 2005 – 2018). The only qualitative indication about the nature of these migrators in the Cretan Sea is one tow made above the ADCP in December 2013, that captured large organisms (\Rightarrow larger than 5 mm) from which known migrators were decapod larvae, euphausiid larvae, siphonophores and chaetognaths (~~Figure 11~~). Indications can also be given by studies targeted on zooplankton migrators in the Western Mediterranean Sea by Andersen and collaborators (Andersen and Nival, 1991; Andersen ~~and Sardou, 1992; Andersen~~ et al., 1992-a & b, 2004; Sardou et al., 1996). Among the several migrant species reported, the most abundant ones that were present all year round (euphausiids, siphonophores and decapods) were concentrated above 150 m at night-time, whereas during day-time the depth of their maximum abundance was found seasonally variable (between 300 m and 500 m) (Sardou et al., 1996). These groups appeared to have similar behavior to group B in the present study. Small euphausiids migrated from 420 m to 240 m, whereas non-migrants remained below 300 m (Sardou et al., 1996), with similar behavior to groups C and A respectively in the present study.

The above work reveals a significant problem associated with the in situ sampling of the above mentioned zooplanktonic groups. Considering that the clear majority of samplings in the area take place during day time, above 100 m depth (when groups A, B, C, D are at the deeper parts) and with inappropriate net type and tow to capture large organisms (as explained above), it is rational to assume that they are misrepresented in the samples. Appropriate sampling strategy with day and night sampling regularly (monthly frequency) with appropriate net type and tows to study diel and seasonal variation of large organisms, has been done in few locations such as the Ligurian Sea (Sardou et al., 1996), the ALOHA site (Al-Mutairi and Landry, 2001) and the BATS site (Madin et al., 2001; Jiang et al., 2007), with significant logistical effort.

4.3 Implications for biogeochemical cycles

If large stocks of large zooplankton actively migrate over significant vertical distances, in an oligotrophic deep system such as the Cretan Sea, then, new carbon pathways will have to be included in our models, reconsidering the energy flow and the dynamics of the system. In fact, since the carbon inflow (feeding) to the migrant groups comes from lower trophic levels (i.e. phytoplankton) at the euphotic zone, the zooplankton migrators may cause an important active downward vertical flux of matter, thus increasing the biological pump's efficiency (review by Frangoulis et al., 2004). The ~~lack~~gaps of ~~data~~ ~~from~~knowledge for midwater depths severely limits our ability to quantify the efficiency of the biological pump (Robinson et al., 2010). In the Cretan Sea, the lack of knowledge of the role of zooplankton ~~vertical-migration~~DVM and the functioning of the whole mesopelagic ecosystem may constitute an important knowledge gap of the biological pump's efficiency in the area that requires exploration. Additionally, the observed patterns are expected to have significant implications in the system dynamics particularly if one considers the oligotrophic character of the Cretan Sea. The observed ~~migration~~DVM is expected to act as a transfer mechanism of organic matter (carbon and nutrients) from the euphotic zone to the deeper parts of the water column, overcoming the physical barrier of the pycnocline. This active flux of matter may occur since the ~~migrations-speed~~

DVM speeds recorded (\Rightarrow larger than 3 cm s^{-1}) were higher than reported zooplankton faecal pellet sinking speeds (\Leftarrow higher than 1 cm s^{-1} for euphausiids - review by Frangoulis et al., 2004). This mechanism will enhance the oligotrophism of the mesopelagic layer since there are no effective mechanisms of very deep-water mixing –and there is a strong decoupling of the surface layers with the deeper parts of the water column. Thus, the surface layers are deprived of important nutrients, although in the actual nutrient budget one has to take into account other parameters such as zooplankton excretions at the surface layers etc.

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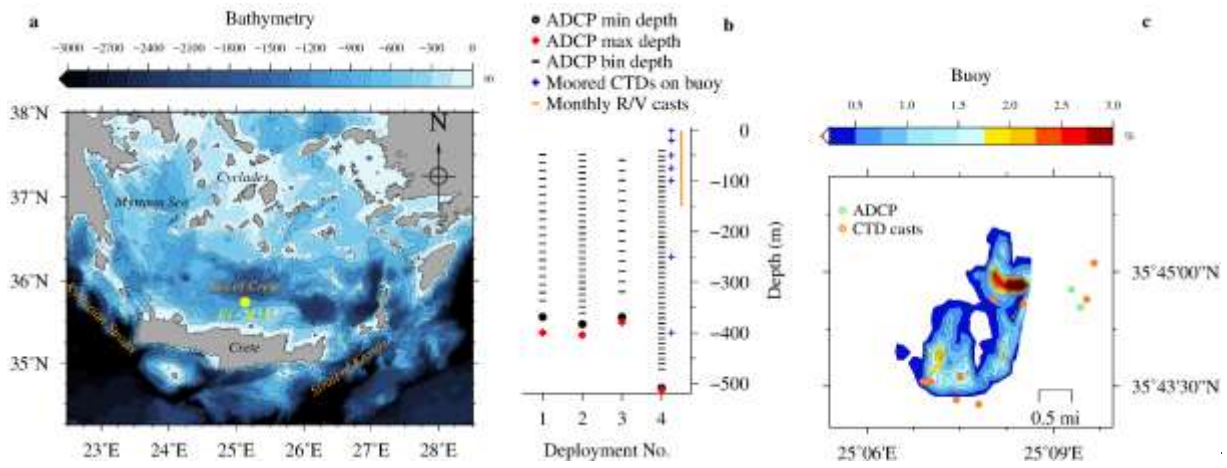
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Table 4-1: The deployment parameters of the upward looking 75 kHz RDI ADCP on the subsurface mooring line of E1-M3A are listed.

Deployment	Start	End	Bins	Bin size (m)	Sampling interval (s)	1 st Bin (m)	Average depth (m)
1 st	15-Nov-2012	23-May-2013	25	16	1800	24.59	369
2 nd	01-Jun-2013	19-Jan-2014	33	12	3600	20.65	383
3 rd	19-Jan-2014	10-Oct-2014	25	20	1800	28.58	370
4 th	10-Oct-2014	02-Jun-2015	45	10	1800	18.76	509

Table 2:2: The type, source, time coverage and resolution of the auxiliary data are listed. In situ data have gaps of variable length. Monitoring by R/V refers to the monthly monitoring program by regular R/V visits at the E1-M3A observatory site. NASA refers to the Goddard Space Flight Center.

Parameter	Type	Source	Time Coverage	Time Resolution
Air temp & wind	<i>In situ</i>	E1-M3A buoy	2013/05-2014/10	3 h
Surface currents (0-50 m)	<i>In situ</i>	E1-M3A buoy (ADCP 400kHz)	2013/05-2015/05	3 h
Subsurface currents (0-400 m)	<i>In situ</i>	ADCP (75kHz)	2012/11-2015/05	0.5-1 h
Water temp & sal	<i>In situ</i>	E1-M3A buoy	2013/05-2015/01	3 h
	<i>In situ</i>	Monitoring by R/V	2010/03-2015/01	1 m
	Reanalysis	SeaDataNet	Climatology	1 m
pH	Reanalysis	SeaDataNet	Climatology	1 m
Chl- α	<i>In situ</i>	E1-M3A buoy	2013/05-2014/06	3 h
	<i>In situ</i>	Monitoring by R/V	2010/03-2015/05	1 m
Cloud fraction & optical thickness	Satellite	NASA	2015/02-2015/03	1 d



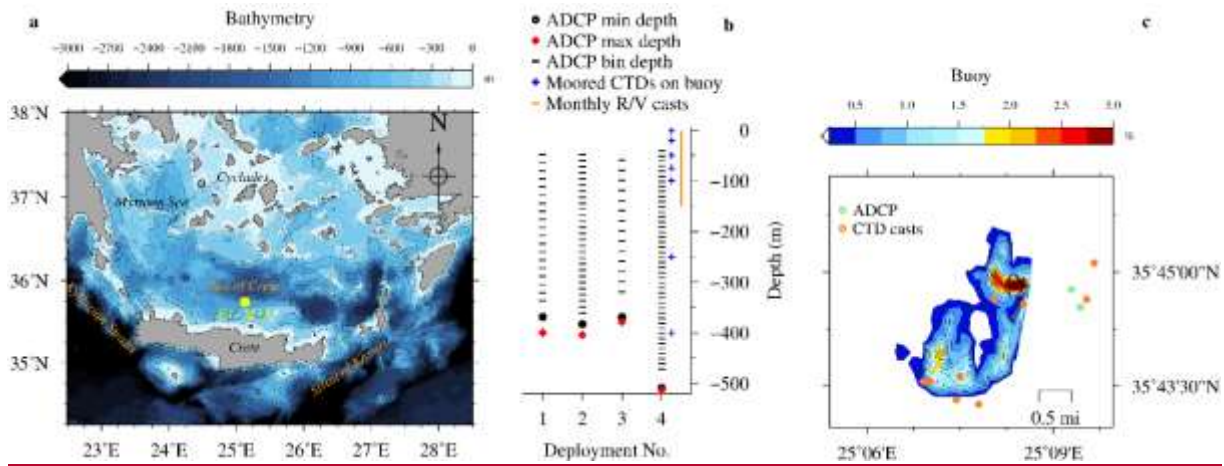
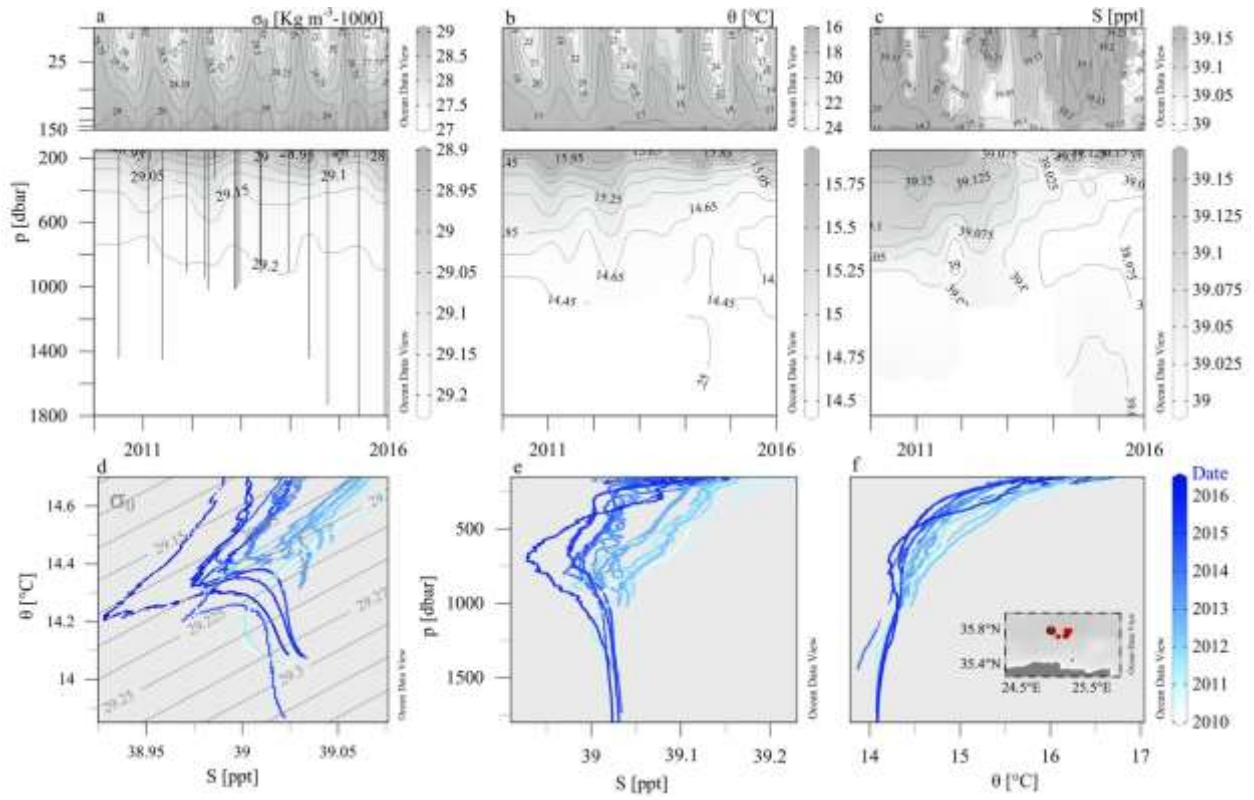


Figure 1: Topographic map of the south Aegean Sea (a), vertical (b) and horizontal (c) views of the sampling set up at E1-M3A. Details of ADCP deployments are given in Table 1. Horizontal buoy motion is shown as the percent of the time of total deployment duration spent at a location.



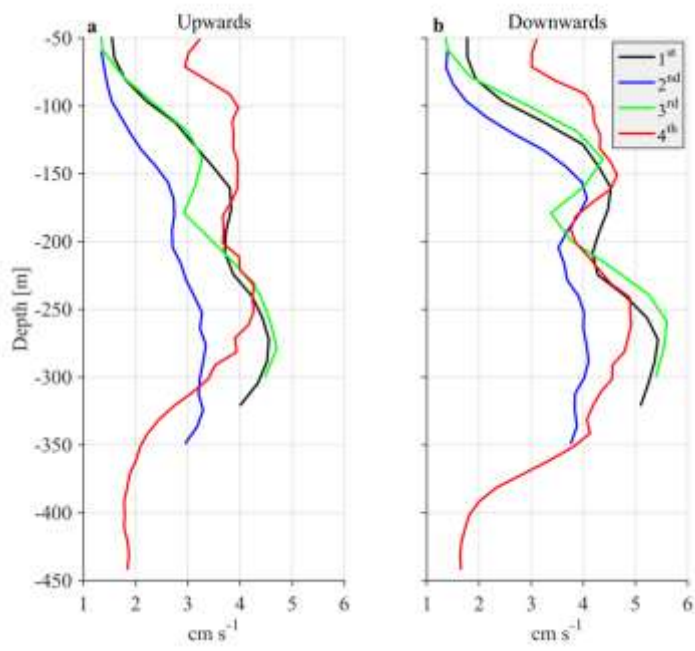


Figure 2: Time average burst speeds per ADCP deployment.

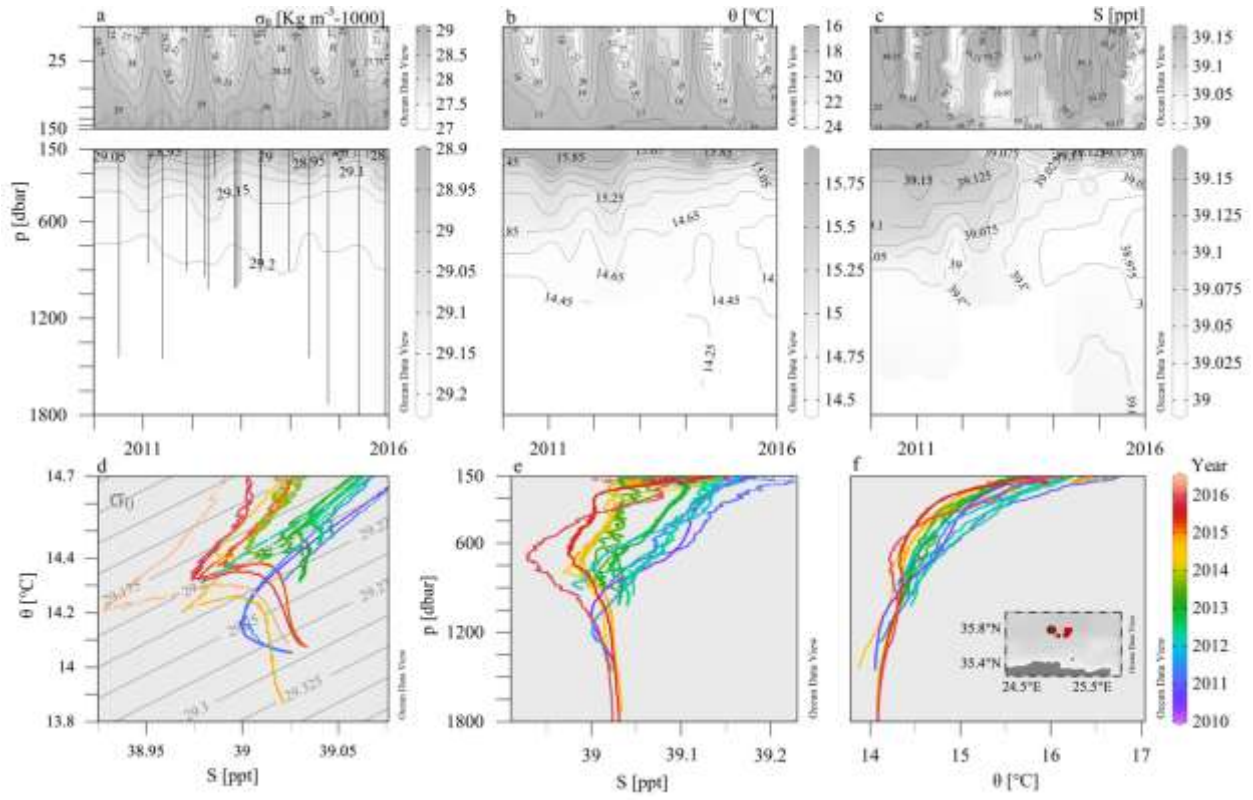
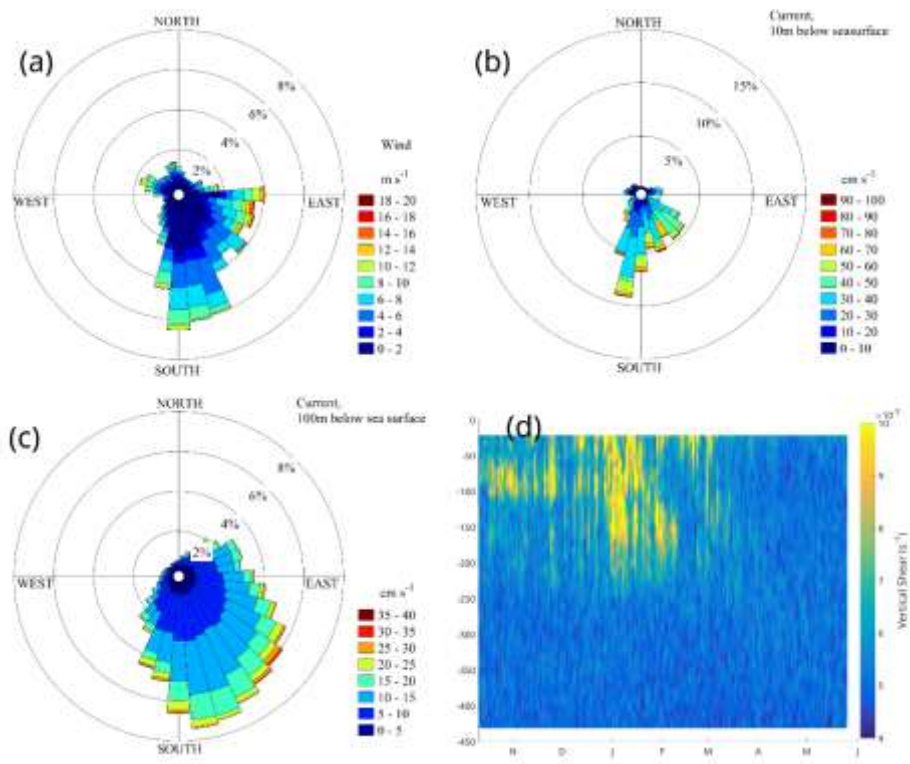


Figure 3: CTD casts collected during monitoring and maintenance visits at the site of E1-M3A observatory with HCMR research vessels *Aegaeo*, *Philia* and *Iolkos* from 2010 to 2016. Time-depth plots of potential density anomaly σ_0 (a), potential temperature θ (b) and practical salinity S (c). $\theta - S$ plot (d) and vertical profiles of S (e) and θ (f) are colored according to date to reveal temporal trends.



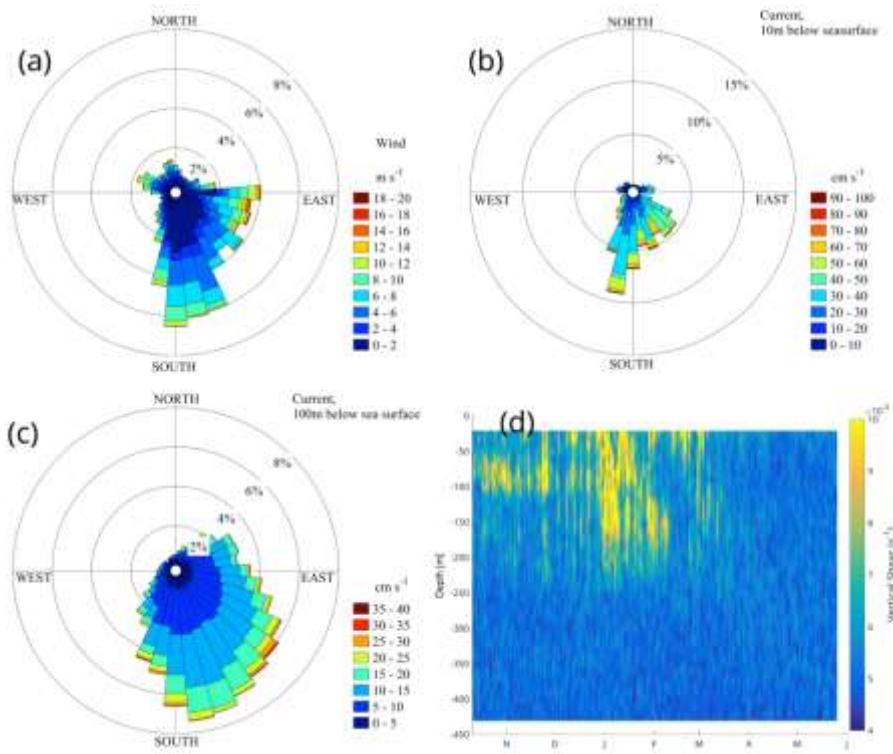


Figure 34: Rose diagrams of wind (a) and surface (10 m depth) currents from the buoy's ADCP (400 kHz downward looking) (b), subsurface currents at 100 m depth from the upward looking 75 kHz ADCP (c) and vertical shear from the fourth deployment (d). The direction in panels (a, b, c) point to the direction of the flow.

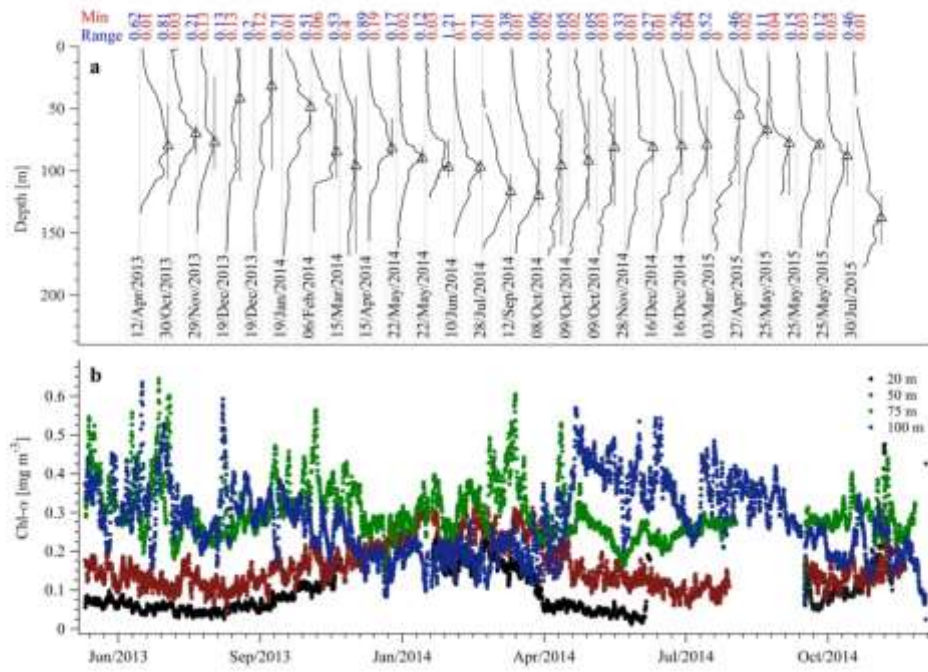


Figure 45: Chlorophyll concentration from the CTD casts (a) and E1-M3A CTD sensors (b). The casts show the vertical distribution of chlorophyll concentration in the water column (normalized, solid black lines). The minimum value of each cast is denoted by the vertical grey-dotted line and the maximum value of each cast is denoted by the black triangle. The gray bars around the triangle denote the depth range for which the chlorophyll concentration is above 70-% of the maximum value of the cast. The minimum value and the range of original chlorophyll values (in mg m^{-3}) are shown above each cast in red and blue colors respectively. E1-M3A chlorophyll data are low passed with a one-day running mean filter.

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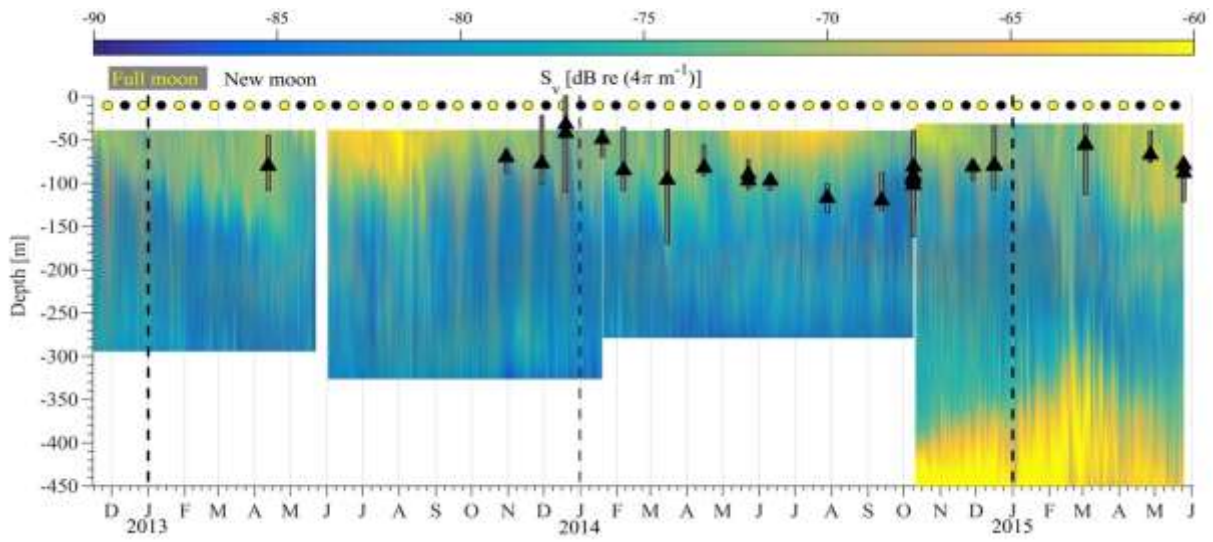


Figure 56: The backscatter coefficient for all ADCP deployments is shown. The beginning of a year is denoted by the dashed vertical line. The ~~black and~~ yellow ~~and black~~ circles denote the dates of full moon and new moon respectively. ~~The~~ The maximum chlorophyll value of available casts is denoted by the black ~~triangles and the~~ triangle. The gray bars have around the same meaning as in Figure 4, ~~at~~ triangles denote the depth range for which the chlorophyll concentration is above 70% of the maximum value of the cast.

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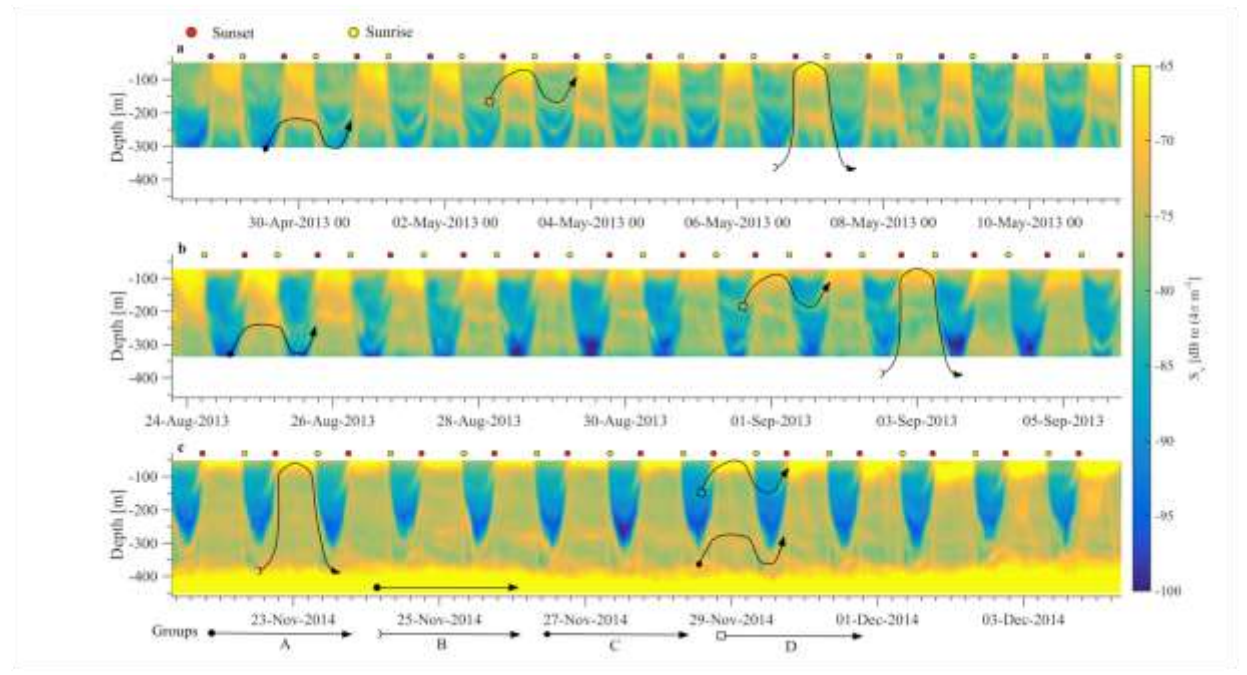
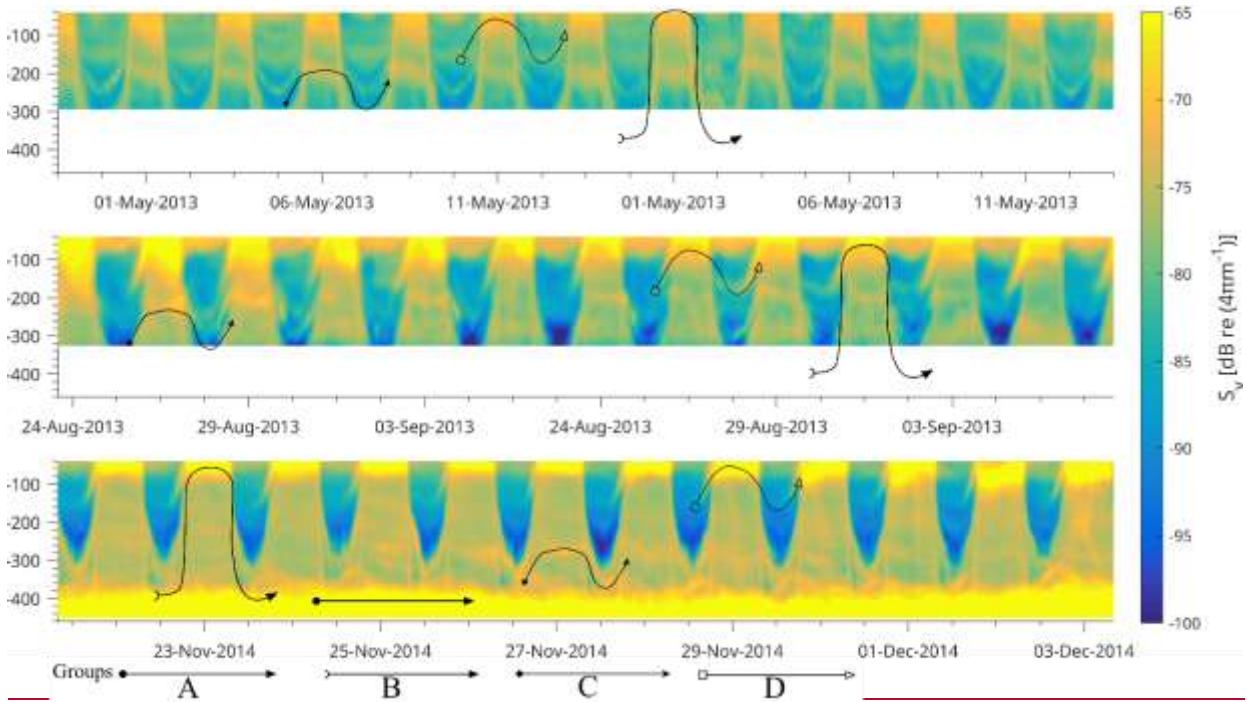


Figure 67: Hand drawn trails of S_v attributed to groups of planktonic and micro-nectonic organisms.

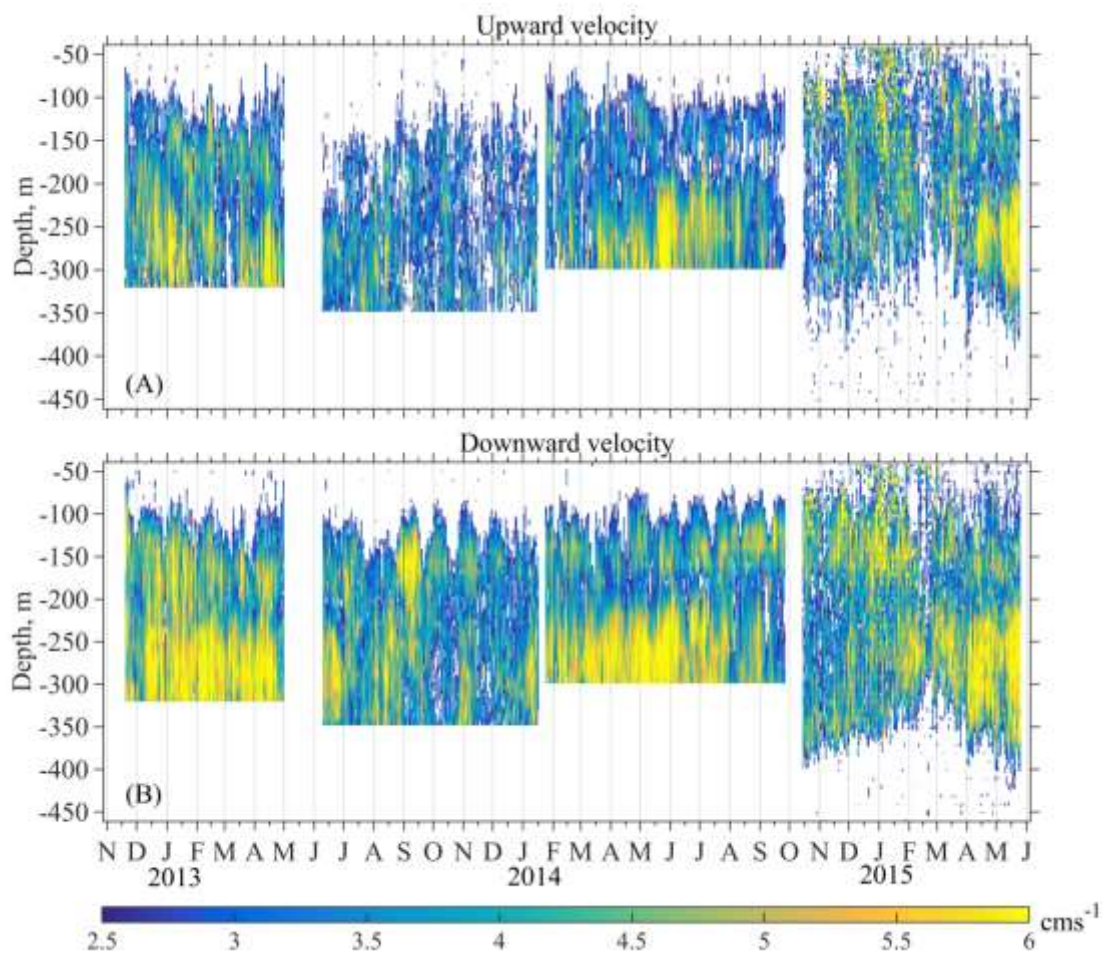


Figure 7: Large upwards (A) and downwards (B) speed, attributed to the migration of zooplankton.

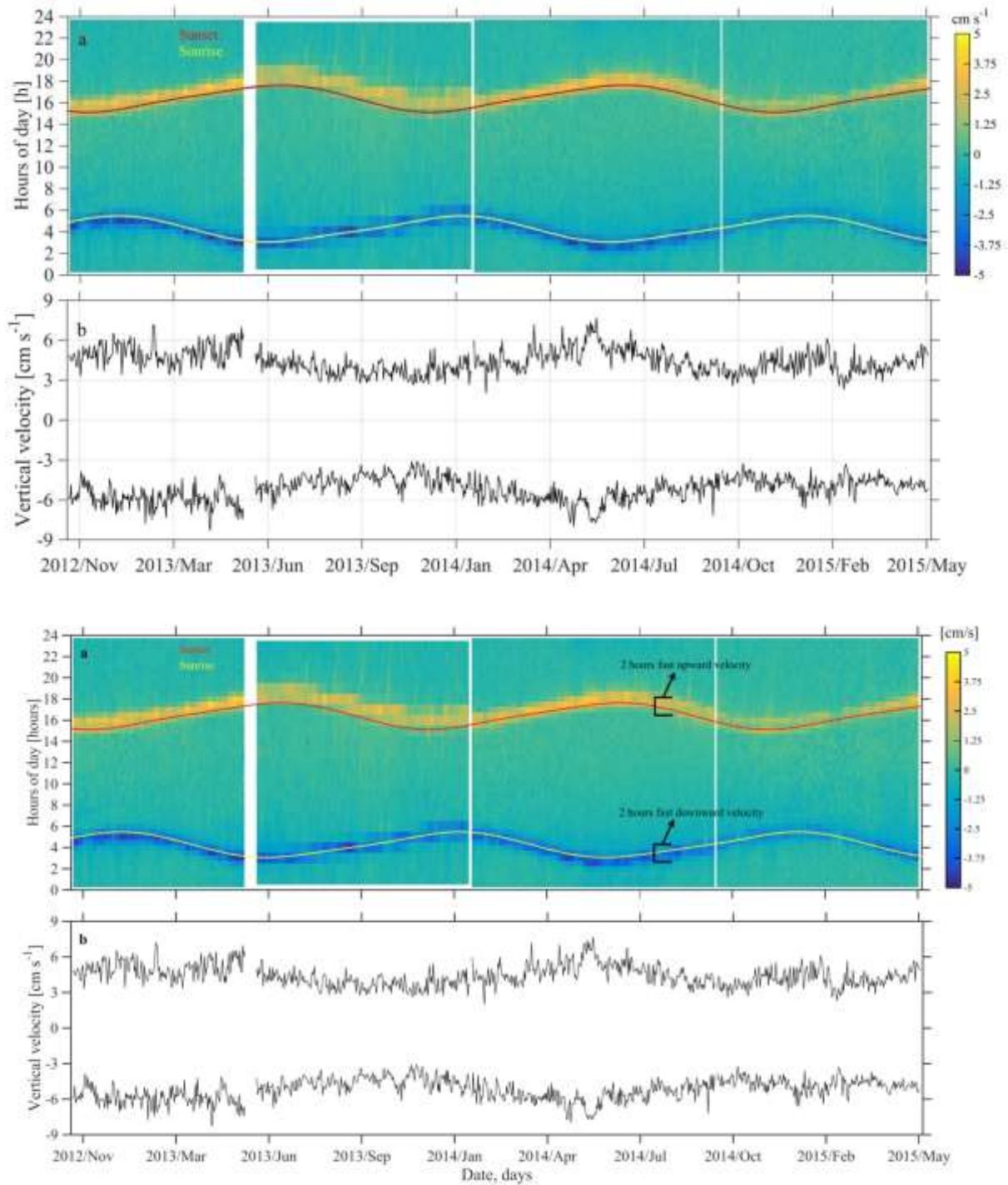


Figure 8: (a) Instantaneous depth averaged vertical velocities of daily segments of ADCP measurements (a) between 350 m and 50 m, following Jiang et al. (2007). ~~Average of the three largest speed measurements per day (b).~~ Sunrise and sunset times are superimposed. (b) Average of the three highest upwards and downwards velocity values per day. The hours of fast zooplankton motion are also shown.

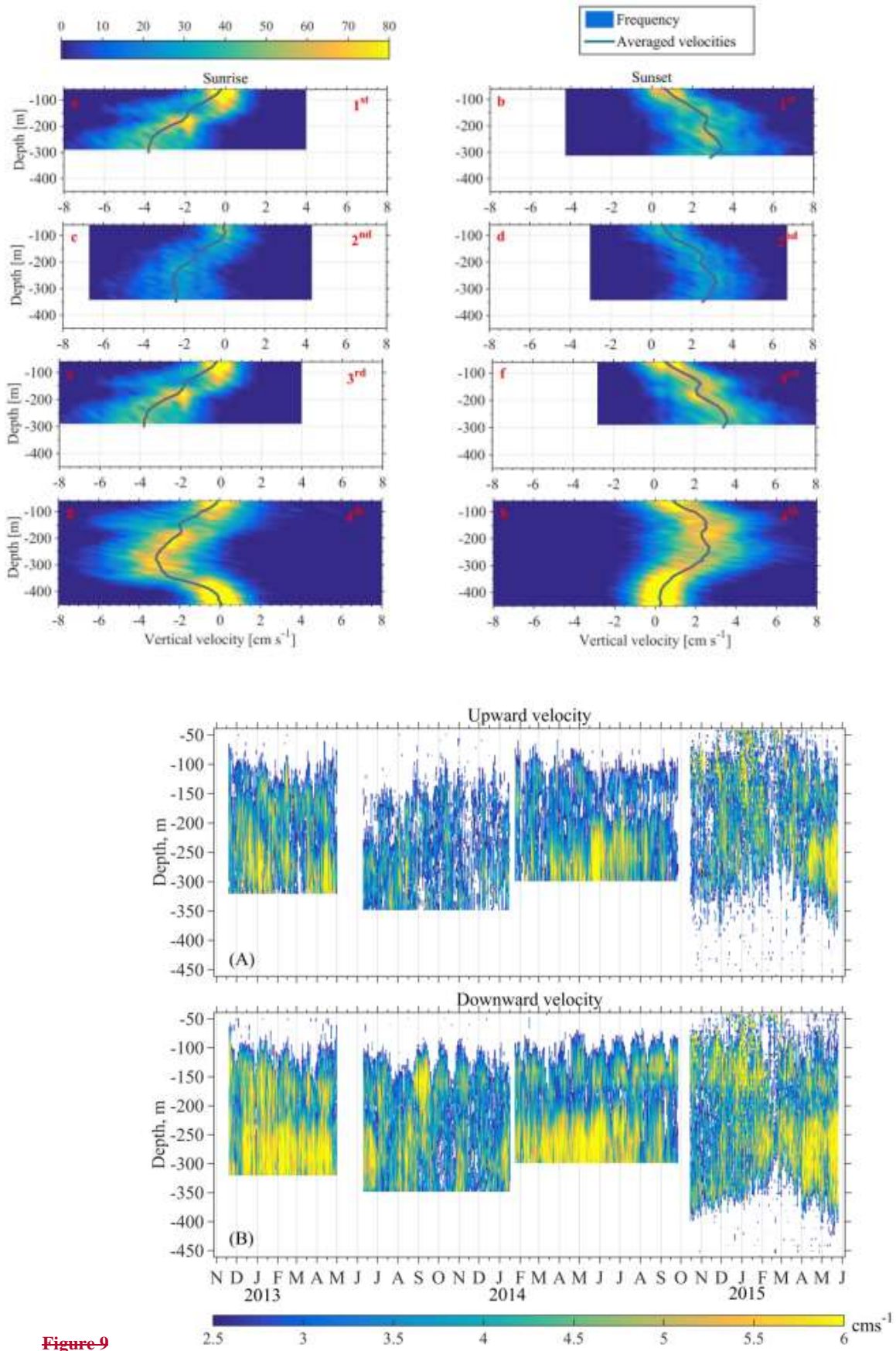


Figure 9

Figure 9: Large upwards (A) and downwards (B) velocity, attributed to the migration of zooplankton.

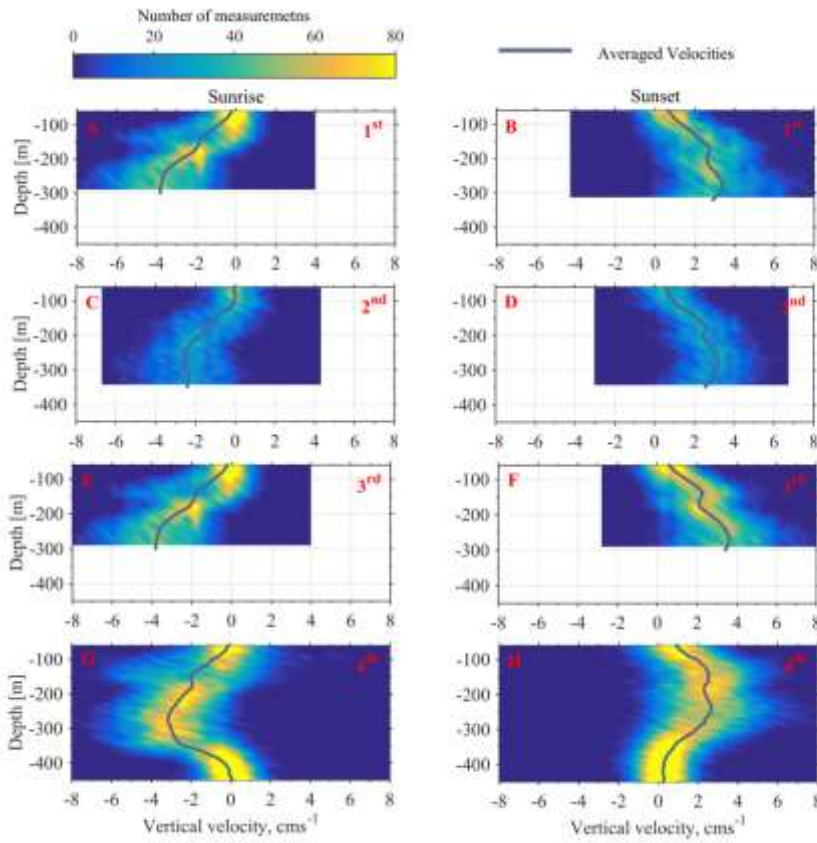


Figure 10: Depth distributions of the vertical velocities, measured 1 h before and 1 h after sunset and sunrise, are shown. The time average velocity at each depth is superimposed. Each row of panels refers to one deployment (1st, 2nd, 3rd, 4th). The first column of panels corresponds to sunrise and the second column to sunset.

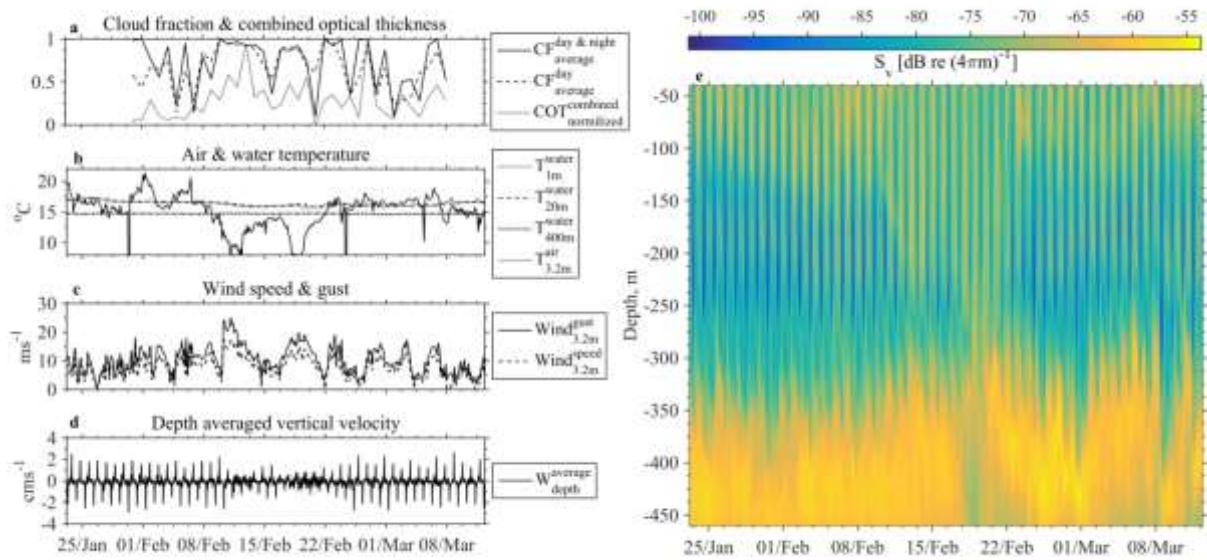


Figure 10

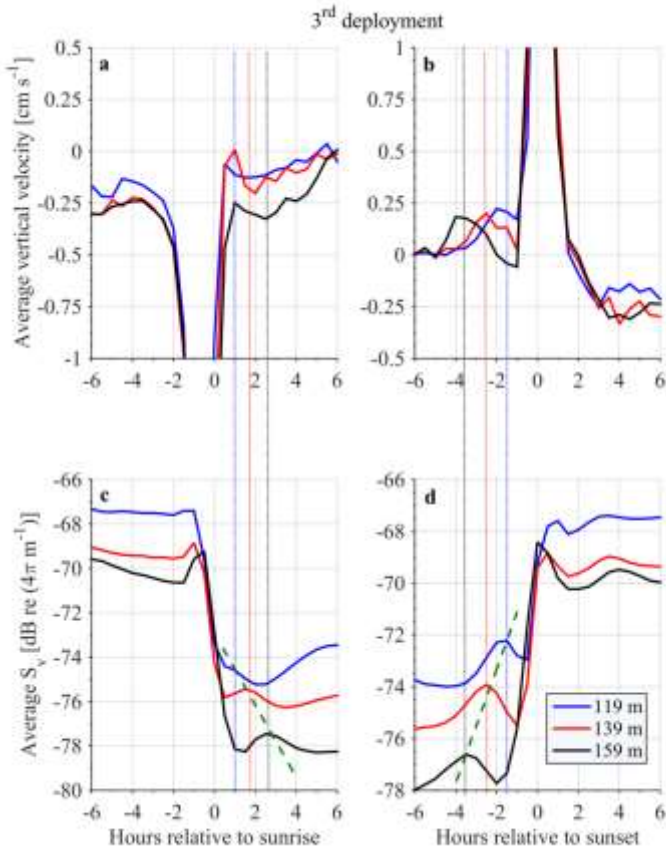


Figure 11: Time average vertical velocity (a, b) and S_v (c, d) at selected depths during the 3rd deployment. The green dashed line connects the S_v peaks attributed to group D. The vertical dotted lines are used to facilitate the common vertical velocity and S_v peaks, attributed to group D.

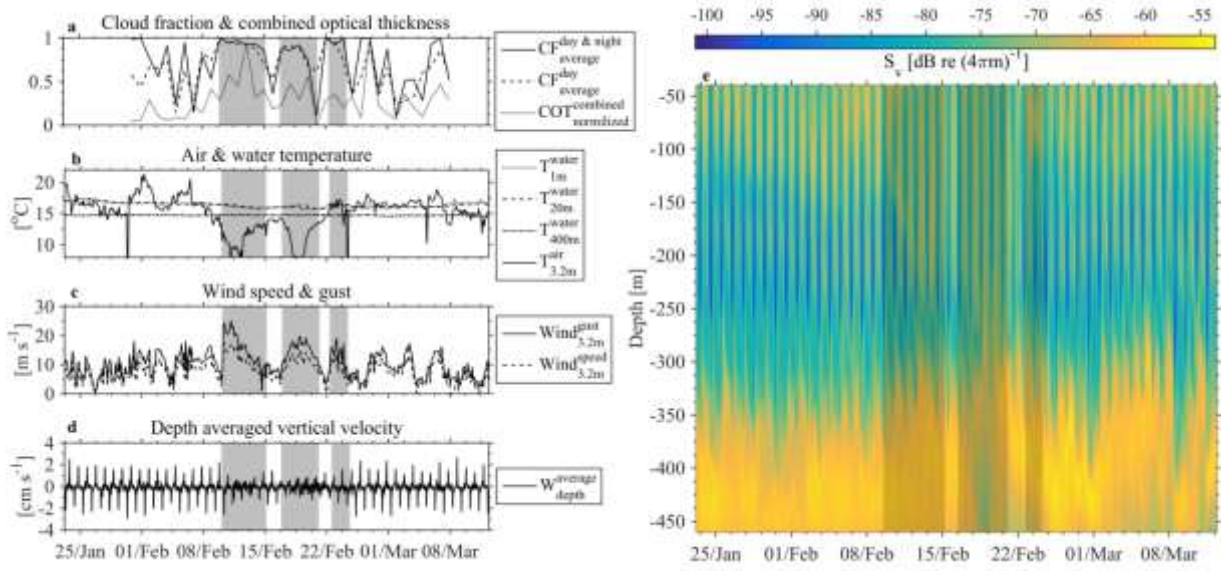


Figure 12: Cloudiness (a), air and water temperature (b) and wind conditions (c) are examined in comparison to depth averaged vertical velocities (d) and backscatter coefficient (e) during February 2015. Gray shaded areas denote the three harsh weather events referred in the text.



Figure 11: Image of the larger organisms captured with a 500 µm mesh-size net in the 0-350 m layer at the E1-M3A location in December 2013 at mid-day (petri dish diameter is 8 cm).

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