Spatial Representivity of Plankton Indicators

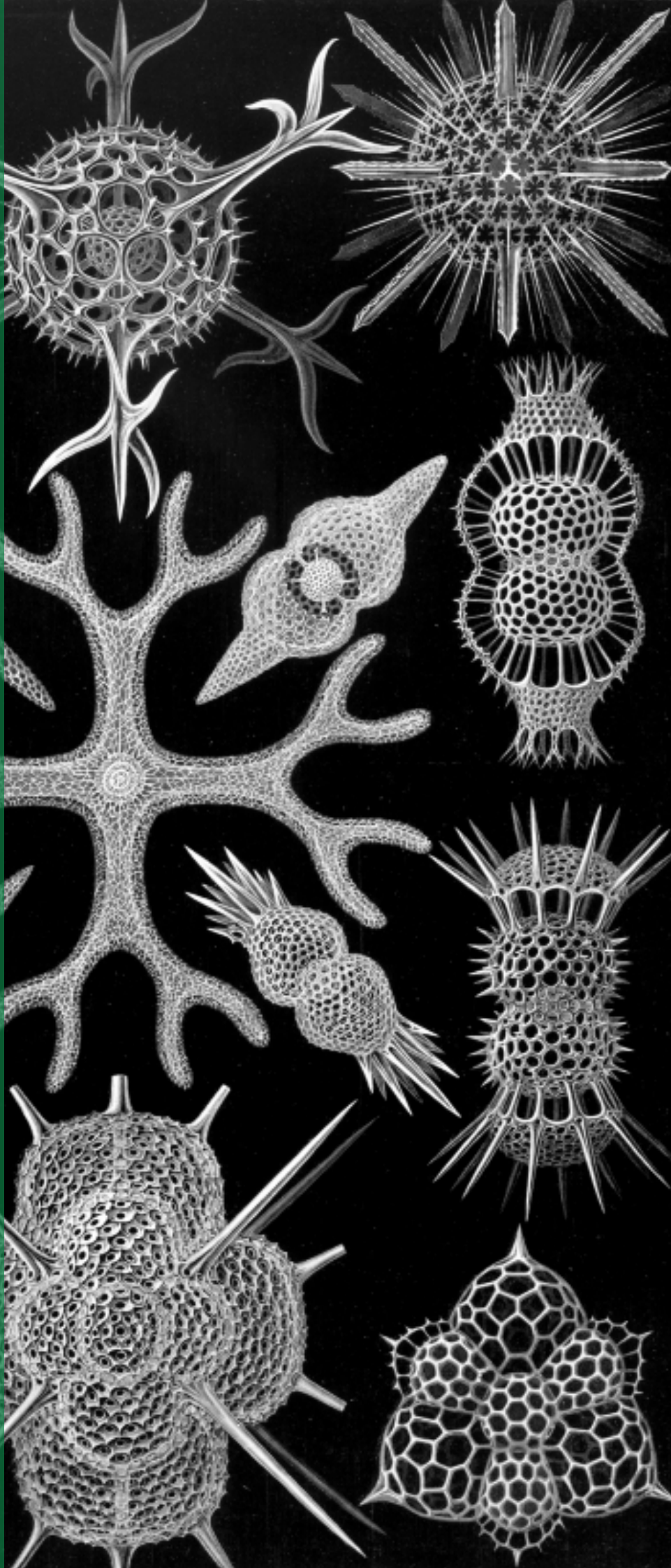
EcApRHA Deliverable WP1.3

Co-financed by the European Union

EcApRHA an Ecosystem Approach to Regional Habitat Assessments

2017

Contents



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EcApRHA

The EcApRHA project (Applying an Ecosystem Approach to (sub) Regional Habitat Assessment) aims to address gaps in the development of biodiversity indicators for the OSPAR Regions. In particular, the project aims to overcome challenges in the development of indicators relating to the MSFD (Marine Strategy Framework Directive 56/2008/EU), such as Descriptor D1 (Biodiversity), D4 (Food webs) and D6 (Seafloor integrity), and to deliver an action plan to OSPAR that will enable monitoring and assessment at the (sub) regional scale, to contribute to OSPAR Intermediate Assessment 2017.

Indicators related to the benthic and pelagic habitats, as well as food webs, are investigated within the project at different levels (from data to indicator; from indicator to habitat assessment; from habitat to ecosystem assessment).

Acknowledgment

This report was produced as a result of the EcApRHA (Addressing gaps in biodiversity indicator development for the OSPAR Region from data to ecosystem assessment: Applying an ecosystem approach to (sub) regional habitat assessments) project. The project was co-financed by the European Union (EU). Grant No. 11.0661/2015/712630/SUB/ENVC.2 OSPAR

Executive summary

This report is part of a suite of actions to support the development of "applying an ecosystem approach to (sub) regional habitat assessments (EcApRHA)" in the North-East Atlantic. This report is centered on increasing our understanding of the spatial representivity of plankton indicators, which was identified as a key knowledge gap in the use of differing plankton datasets, responding to key issue 1 (from data to indicators) and 2 (from indicators to habitat (structure) assessment). This is an important knowledge gap as we are working towards understanding how much of an area a sample represents, which is necessary in order to form regional assessments that are spatially robust. Data from a fixed-point station (L4, located within the Western English Channel) were compared with transect data collected by the Continuous Plankton Recorder (CPR) within extending zones around L4, for two of the Pelagic indicators (PH1 (changes in plankton functional types (lifeform) index ratio) and PH2 (changes in plankton biomass/and or abundance)). This preliminary investigation into the spatial representivity of the outputs from PH1 and PH2 finds that the 'patchy' distribution of plankton limits the ability to merge these differing datasets and compare them inter-annually. However, strong similarities are found between seasonal means and long-term trends, suggesting that there is potential to 'gap-fill' using both forms of data. To accurately define the spatial representivity of a fixed-point station, more information is required on the physical conditions surrounding the station and further analyses of individual taxa and their range in relation to those physical conditions is necessary. It is clear that both forms of data provide useful and complimentary information on the plankton, with fixed-point data providing detailed information on localized conditions, while CPR data provides critical information on regional conditions and long-term trends. Where possible, both forms of data should be used for providing a more complete story of the conditions of the pelagic habitat.

1 Introduction/Background

The project "applying an ecosystem approach to (sub) regional habitat assessments (EcApRHA)" is centered on addressing gaps in the application of biodiversity indicators in the North East Atlantic. These gaps have been focused into actions in order to aid the development of such indicators, which are being developed within the OSPAR Commission[1] in response to the Marine Strategy Framework Directive (MSFD) and support the assessment of moving towards Good Environmental Status (GES). This deliverable is focused around action 1.3 of the project, which addresses the need to understand the spatial representivity of plankton indicators. There are two differing but complementary forms of plankton data currently available for PH1 (changes in plankton functional types (lifeform) index ratio), PH2 (changes in plankton biomass/and or abundance), and PH3 (changes in biodiversity indices); fixed-point data, and transect data. There are three key differences between the two data sources for the net-based estimates. First the replicate pair of nets used weekly at the L4 monitoring station sample nearly the whole water column (vertically from 50 m to the surface in a ~54 m water column) during daytime, whereas the CPR nets integrate the near-surface wash of the propeller (for example the top 10 m layer) and in both day and night. Second the CPR is 270 um mesh, substantially greater than the 200 um mesh of the plankton net that is used to sample at L4 (WP2 net). Thirdly, L4 sampling is at the fixed site only 13 km from Plymouth; this is near the inshore limit of the CPR data coverage, and due to the blocking nature of the land (Fig 1), the L4 is likely to have an inshore bias compared to CPR. Richardson *et al.* (2006) provide detailed CPR methodology and Widdicombe *et al.* (2015) and Atkinson *et al.* (2015) describe methods for Chl *a*, microplankton and zooplankton at L4. The CPR survey is coordinated by the Sir Alister Hardy Foundation for Ocean Science (SAFHOS) in the UK, and has tows within the English Channel and within close proximity to L4.

There have been a number of studies that have compared plankton data between L4 and CPR data, with broad conclusions suggesting that although the CPR tends to under-sample plankton abundance relative to fixed-point data, the general seasonal and long-terms trends are similar (John *et al.* (2001), Scheef *et al.* (2012), Owens *et al.* (2013)). Scheef *et al.* (2012) compared the output from a multivariate autoregressive model to investigate community structure using L4 data and CPR data from increasing zones around L4. They found no correlation between the outputs from the two different datasets; however, the CPR data in closest proximity to L4 gave the most similarly matched results. Scheef *et al.* (2012) concluded that the lack of correlation between the datasets was due to a combination of observation error and spatial variation in plankton (patchiness). This report uses a similar method of extending zones around L4 to compare the two differing datasets using the outputs from both PH1 and PH2.

2 Aims/Rationale

This study aims to assess the spatial representivity of plankton data using the outputs from both PH1 and PH2. This is a needed step in understanding of how representative a fixed-point station is of a spatial area. Following on from the aggregation of disparate fixed-point (local scale) and transect (regional scale) data through the EcApRHA project deliverable 1.1 (Ostle *et al.,* 2017). L4 (local) and CPR (regional) data shall be used to determine if both data types show similar findings. The potential for use of a 'conversion factor' shall also be investigated, in order to extrapolate and 'gap fill' where data may be missing. In order to make regional assessments using indicators, it is

[1] OSPAR is the mechanism by which 15 Governments & the EU cooperate to protect the marine environment of the North-East Atlantic. www.ospar.org

important to understand how much of an area samples used within the assessments represents. This study uses a case-study location within the English Channel to initiate these investigations, and suggest potential next-steps to achieve spatial robustness within analyses.

3 Methodology

3.1 Sample locations, and spatial aggregation

The L4 monitoring station was chosen as the study site in order to investigate the spatial representivity of the plankton indicators because it has a consistent time series and CPR samples within close proximity. In order to assess the spatial representivity of the datasets, CPR samples were averaged from extending zones around L4 (see **Figure 1**). To make sure that no CPR samples were aggregated from seas outside of the Western English Channel, the CPR samples used within this analysis were restricted to within the black polygon shown in **Figure 1**.

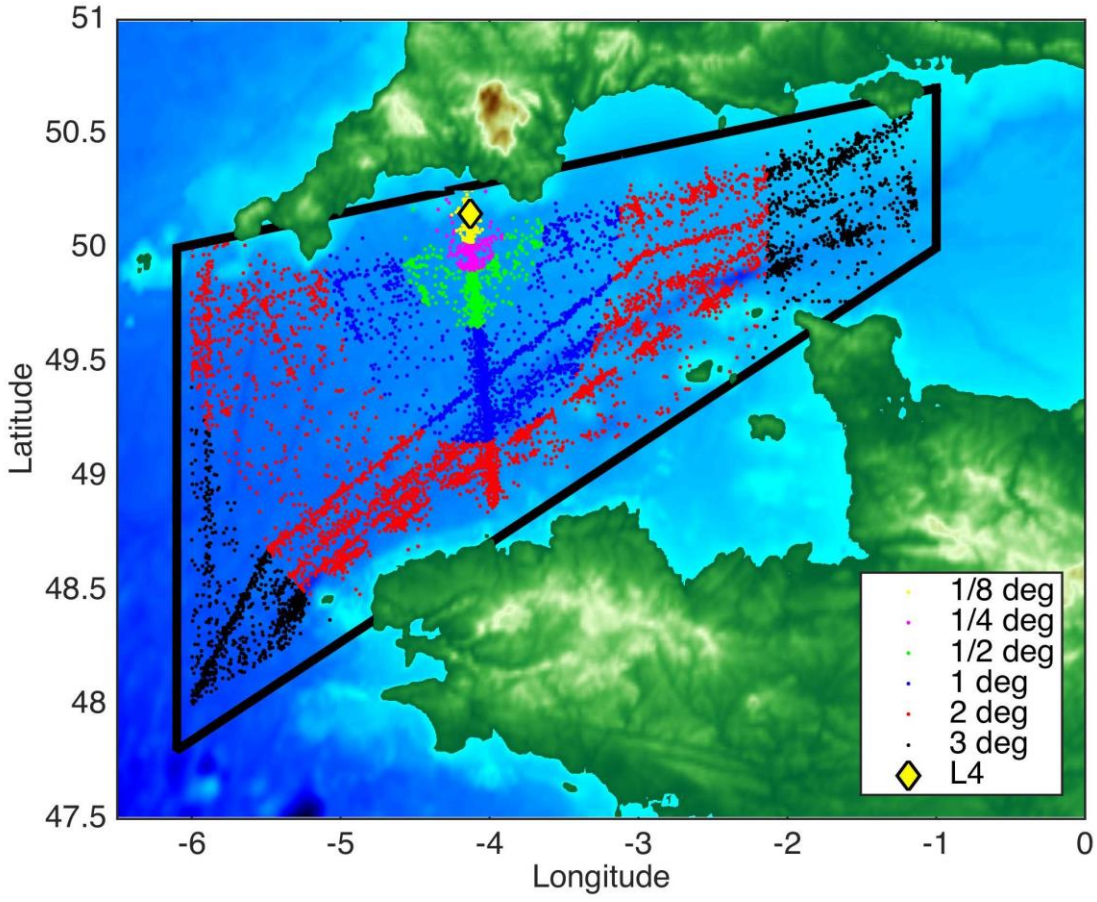

Figure 1: Map of the sample locations. The coloured dots represent CPR samples from extending ranges around L4, which is represented as a yellow diamond. Cyan dots = CPR samples within 1/8 degree of L4. Pink dots = CPR samples within ¼ degree of L4. Green dots = CPR samples within ½ degree of L4. Dark blue dots = CPR samples within 1 degree of L4. Red dots = CPR samples within 2 degrees of L4. Black dots = CPR samples within 3 degrees of L4. The areas include all sample points within the range stated, therefore encompassing any ranges that are smaller. The black polygon represents extent of study area used.

3.2 Brief methodology for PH1 Changes in plankton communities.

PH1 "Changes in plankton communities" features a "Plankton Index" of lifeform pairs, which have been developed to track changes in the state of the plankton in marine waters over time. The main features of the method are: (i) the grouping of planktonic species into functional types or lifeforms; (ii) the display of changes in the abundance of each of these lifeforms using a state-space approach; (iii) calculating a Plankton Index (PI) to quantify possible changes in the state of the plankton relative to baseline or starting conditions; and (iv) relating trends in the PI to trends in human pressures and climate change indices. When examined in ecologically-relevant pairs, lifeforms can provide an indication of changes in: the transfer of energy from primary to secondary producers (changes in phytoplankton and zooplankton); the pathway of energy flow and top predators (changes in gelatinous zooplankton and fish larvae); benthic/pelagic coupling (changes in holoplankton (fully planktonic) and meroplankton (only part of the lifecycle is planktonic, the remainder is benthic)) (**Table 1**; see also Gowen *et al.,* 2011). For the spatial analysis between L4 and CPR data, the monthly lifeforms were calculated for CPR data within each spatial range shown in **Figure 1** and the annual PI timeseries were compared with L4 for the period 2004 to 2014 (with a reference period of 2004 to 2008). Detailed methodologies for this indicator, and the datasets used, are provided for the PH1/FW5 indicator assessment sheet within the OSPAR Intermediate Assessment 2017 (OSPAR, In prep).

Using expert opinion, each lifeform was evaluated on two characteristics: the ability to identify and speciate organisms in that lifeform using light microscopy and the understanding of the accuracy of determining traits assigned to the lifeform. For example, low confidence is assigned to the lifeform pair 'diatoms vs auto- and mixo-trophic dinoflagellates' as the mixotrophic and autotrophic mode of feeding of many dinoflagellates species is currently uncertain. Thus the accuracy of assigning the life form category is low in this case. Likewise, the lifeform pair 'carnivorous zooplankton v. non-carnivorous zooplankton' has a low confidence designation since the feeding habits of many abundant and common zooplankton species remain unknown. Only pairs with two high-confidence lifeforms (8 out of 12 lifeform pairs) were used in the OSPAR reporting (**Table 1**).

Lifeforms	**Additional criteria**	**Confidence**	**Explanation**
Diatoms v. dinoflagellates		High	Dominance by dinoflagellates may be an indicator of eutrophication or of change in water column stability and may result in less desirable food webs
Gelatinous zooplankton v. fish Larvae/eggs	Ctenophores and cnidaria	High	Indicator of energy flow and possible trophic pathways
Small copepods v. large copepods	Adults <1.9 mm (not nauplii or eggs)	High	Size based indicator of food web structure and energy flows

Lifeforms	Additional criteria	Confidence	Explanation
	Adults >2 mm		
Carnivorous zooplankton v. non-carnivorous zooplankton		Low	Indicator of energy flow and balance between primary consumers and secondary consumers
Crustaceans v. gelatinous zooplankton		High	Indicator of energy flow and possible trophic pathways
Large microphytoplankton v. small microphytoplankton	>20 □m cells, not colonies.	High	Size-based indicator of the efficiency of energy flow to higher trophic levels
	<20 □m cells, not colonies.		
Microphytoplankton v. non-carnivorous zooplankton	Biomass (example Chl, PCI)	High	Indicator of energy flow and balance between primary producers and primary consumers
	Abundance		
Diatoms v. autotrophic and mixotrophic dinoflagellates		Low	Shift in primary producers may indicate eutrophication
Pelagic diatoms v. tychopelagic diatoms		High	Indicator of benthic disturbance and frequency of resuspension events
Nuisance and/or		Low	Shift in algal community towards nuisance

Lifeforms	Additional criteria	Confidence	Explanation
toxin-producing diatoms v. diatoms Or Nuisance and/or toxin-producing dinos v. dinos			and/or toxic species which have the potential to impact other higher trophic level indicators
Holoplankton v. meroplankton		High	Indicator of strength of benthic-pelagic coupling and reproductive output of benthic versus pelagic faunas
Ciliates v. microflagellates	Including tintinnids	Low	Shift from primarily autotrophic to a more heterotrophic system
	All species < 20 µm		

Table 1: Lifeform pairs consist of two ecologically-relevant lifeforms. The 'Additional criteria' column contains supplementary information regarding particular lifeforms.

3.3 Brief methodology for PH2 Plankton biomass and/or abundance

Detailed methodologies for this indicator are provided in the PH2 assessment sheet within the OSPAR Intermediate Assessment 2017 (OSPAR, In prep) and CEMP guidelines (OSPAR, in prep). The basis of the calculation for this indicator is the consideration of the total phytoplankton biomass and the total copepod abundance.

For total phytoplankton biomass, total chl *a* is used for the L4 station. At L4, Chl *a* has been measured through two different methods, fluorometry and HPLC (High Performance Liquid Chromatography) analysis. The data provided from March 1999, were analysed via HPLC analysis, which is a more accurate method to derive chl *a* than the previously used fluorometry. Chl *a* has been almost exclusively determined for surface waters, in contrary for other depth layers. Accordingly, we selected only surface chl *a* (however, due to a gap in measurements of surface chl *a* of more than 4 months in 2011 (from July to December), the chl *a* measured at 10 m depth has been used instead). Also, since the PH2 analysis requires that the time-series start on the month of January for the first year, and that the first value in the year 2000 corresponds to February, we estimated a value of January 2000, assigning it a value equal to the average chl *a* calculated for the whole time-series for the month of January.

For zooplankton, abundances of all copepod species provided for L4 have been summed (except for two taxa) to calculate monthly values of total copepod abundances. Copepod nauplii stages were removed from the total copepod calculation because these stages exhibit high temporal variation.

This may reflect the fact that nauplii are inadequately sampled by the WP2 net (this is a net that is commonly used to sample plankton, 200 um = mesh size), due to net mesh selection, which induces high bias in the estimation of their abundances. One taxon reported in the zooplankton data set, *Diaixis hibernica*, has also been removed from the total copepod abundance calculation since this species is benthic and not planktonic.

For comparison with the CPR dataset, total phytoplankton biomass was estimated based on the Phytoplankton Colour Index (PCI) (Richardson *et al.,* (2006), and total copepod abundance was determined in a similar way to the above but using CPR counts. Although the CPR dataset runs with consistent counts from 1958, zooplankton weekly sampling data at L4 was only from March 1988, and the chl *a* was measured at L4 with HPLC since March 1999. Therefore for the comparison between L4 and CPR data, the total phytoplankton time series runs from 1999 to 2014, and the total copepod abundance time series runs from 1988 to 2014.

Data for total phytoplankton biomass and total copepod abundance were averaged to calculate monthly means of each. All abundance counts were log transformed $\log_{10}(x + 1)$, and the chl *a* and PCI values were left as is, with chl *a* being recorded as mg m^{-3} and PCI not having a defined unit. The anomalies were calculated following the methodology used in the time-series R package Pastecs by F. Ibanez and P. Grosjean (2006). In simplified terms, this is a process of de-seasonalizing the monthly data using the monthly mean cycle, and calculating the annual average anomaly from the monthly anomalies.

4 Results

4.1 Sample size and time-series

Initially when deciding on the spatial range, a smaller radius of 1/16 degrees from L4 was used, however there were not enough samples within the analysis period to justify using such a range. This can be seen from **Figure 2**, where there are < 2 CPR samples for the majority of the analysis period at the smaller spatial range of 1/8 degree radius from L4. There is no optimum sample size defined for monthly aggregation and use with plankton indicators, however it has been suggested that for biodiversity indicators a sample size of >10 is required. This is very dependent on the question at hand, for monthly CPR sample aggregation at 1 degree by 1 degree spatial scale, ≥3 samples has been used in previous studies investigating PCI and copepod abundance (Helaouët *et al.,* 2013).

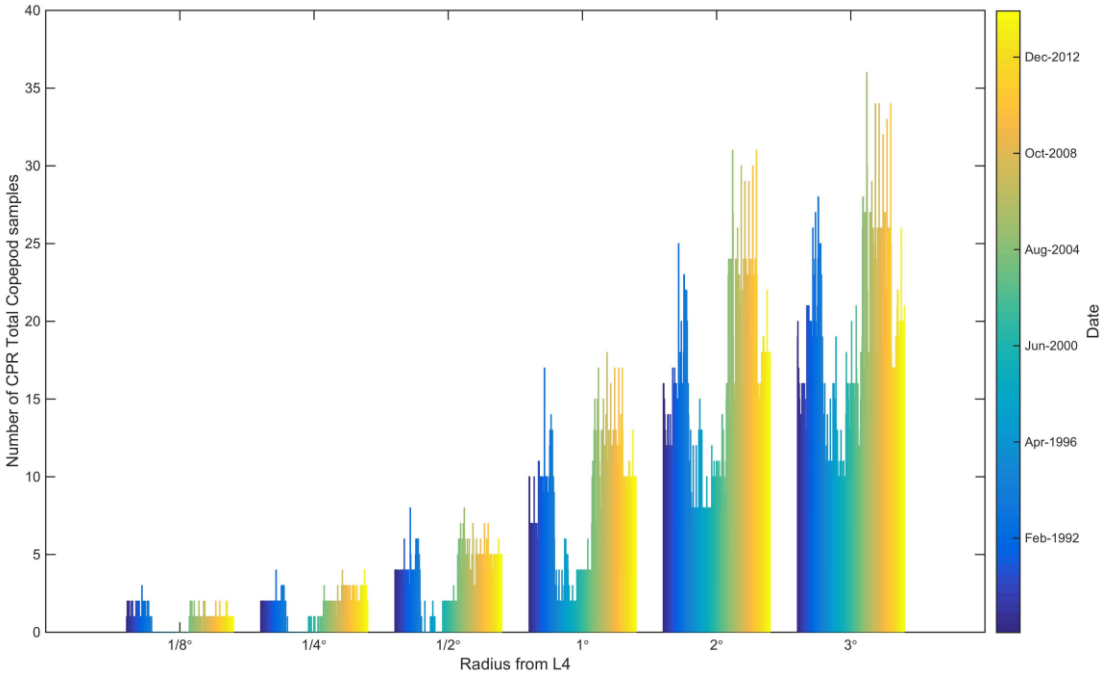

Figure 2: Number of total copepod samples from the CPR within each spatial range of L4 (see **Figure 1**). The colour bar indicates the date at which the sample was collected; the dataset presented is from 1988 to 2014.

There is a clear gap in the number of CPR samples collected between 1995 and 2000, this can be seen in the monthly time series plot of the total copepod abundance in **Figure 3.** This is due to a change in the CPR towed routes during this period, which meant that no CPR samples were collected in close proximity to L4 during this time. Although both datasets are reporting total copepod abundance, the L4 time-series has a greater overall abundance than the CPR time-series (**Figure 3**). This is likely due to the differing sampling methods used; the L4 time-series uses WP-2 zooplankton nets, with a mesh size of 200 µm, while the CPR uses a very different mechanism with a mesh size of 270 µm. Therefore the CPR is likely to capture fewer copepods than the WP-2 nets. It has also been suggested that 'passive avoidance' leads to lower abundance counts in the CPR compared to data from L4 (John *et al.*, 2001).

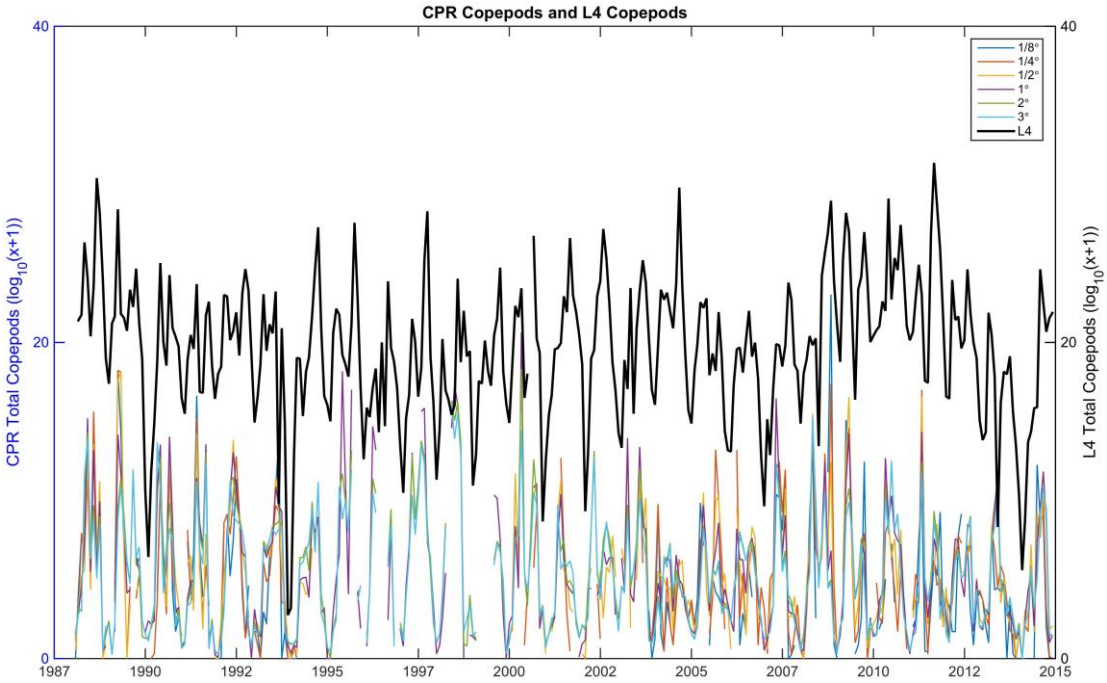

Figure 3: Monthly total copepod abundance ($log_{10}(x+1)$) for L4 (black line) and CPR samples within different spatial ranges of L4 (coloured lines).

Figure 4 shows the monthly time series of chl *a* at L4 and the monthly PCI values at each spatial range around L4. Although the methods are very different from each other they show a similar range in values, and often similar seasonal cycle, with a few exceptions (for example the large peak in chl *a* in 2008 at L4 is not picked up in the CPR PCI samples).

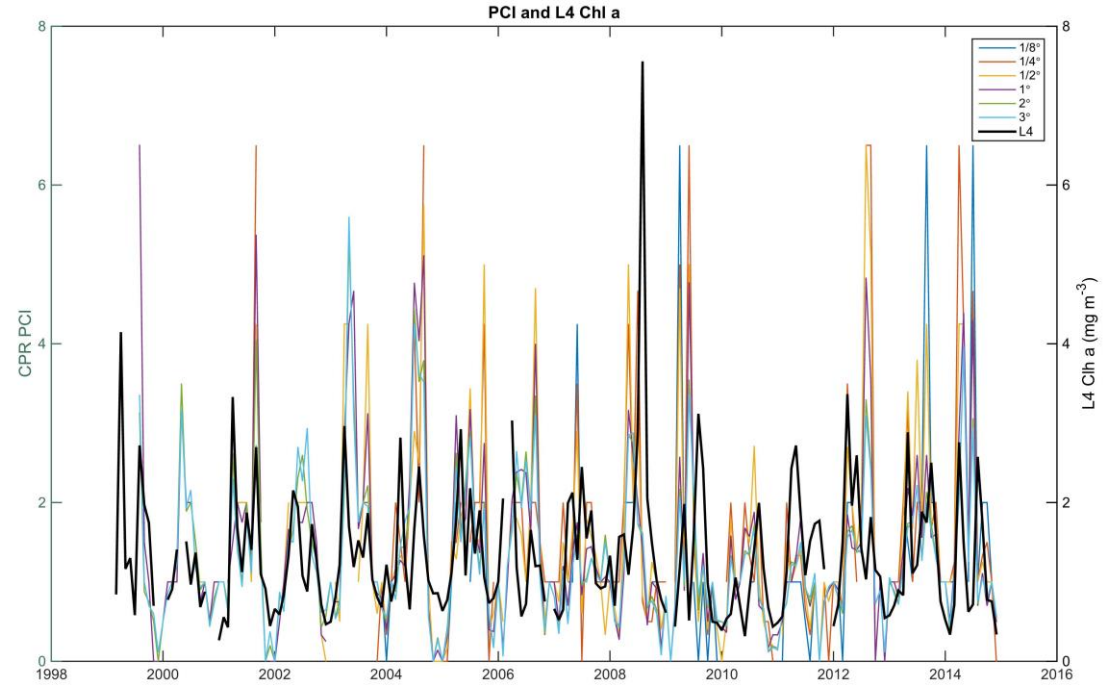

Figure 4: Monthly chl *a* (mg m^{-3}) for L4 (black line) and CPR PCI within different spatial ranges of L4 (coloured lines).

4.2 Conversion factor

A conversion factor is applied here to convert between the two differing datasets in order to fill in gaps of missing data. Pearson's correlation was used to investigate the correlation between the L4 total copepod abundance and total phytoplankton at the different spatial scales of CPR data shown in **Figure 1**. Following Raitsos *et al.*, (2014), the significance of the Pearson's correlation coefficients (r) was adjusted for temporal autocorrelation (pAC) via the effective sample size utilizing the modified Chelton method recommended by Pyper and Peterman (1998). Total copepod abundance from L4 and CPR data showed a significant relationship (pAC < 0.01), with CPR data within ¼ degrees of L4 giving the strongest significance (**Figure 5**).

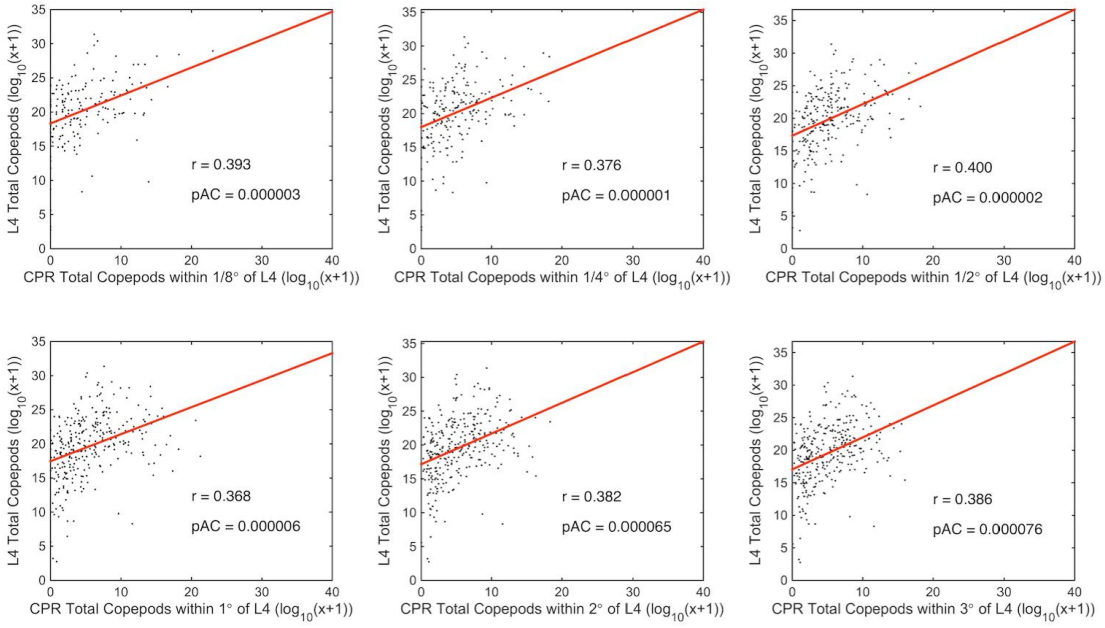

Figure 5: Relationships between total copepod abundance from L4 and the CPR dataset at different spatial scales. Pearson's correlation coefficient is displayed for each relationship as r, and the significance corrected for temporal autocorrelation as pAC.

The conversion factor is calculated using the trend (gradient) and offset (y-intercept) of the relationship giving the most significance (within ¼ degrees). The L4 data can then be used to fill gaps in the CPR time-series (or *vice versa*):

L4_adjusted_to_CPR=(L4_Copepods x trend)+offset

L4_adjusted_to_CPR =(L4_Copepods x 0.3254) -1.4676

This is shown in **Figure 6** with the CPR data shown in blue and the adjusted L4 data shown in red. Although the range of total copepod abundance is not reflected in the adjusted data, the interannual trends are apparent from both datasets, with both showing a similar mean and decrease in total copepod abundance from 2012 onwards. The similarities in long-term trends between the two different datasets are supported by previous findings; such as that of John *et al.*, (2001) who analysed locally abundant copepod species from both L4 and CPR and found they had similar long-term trends as well as seasonal cycles.

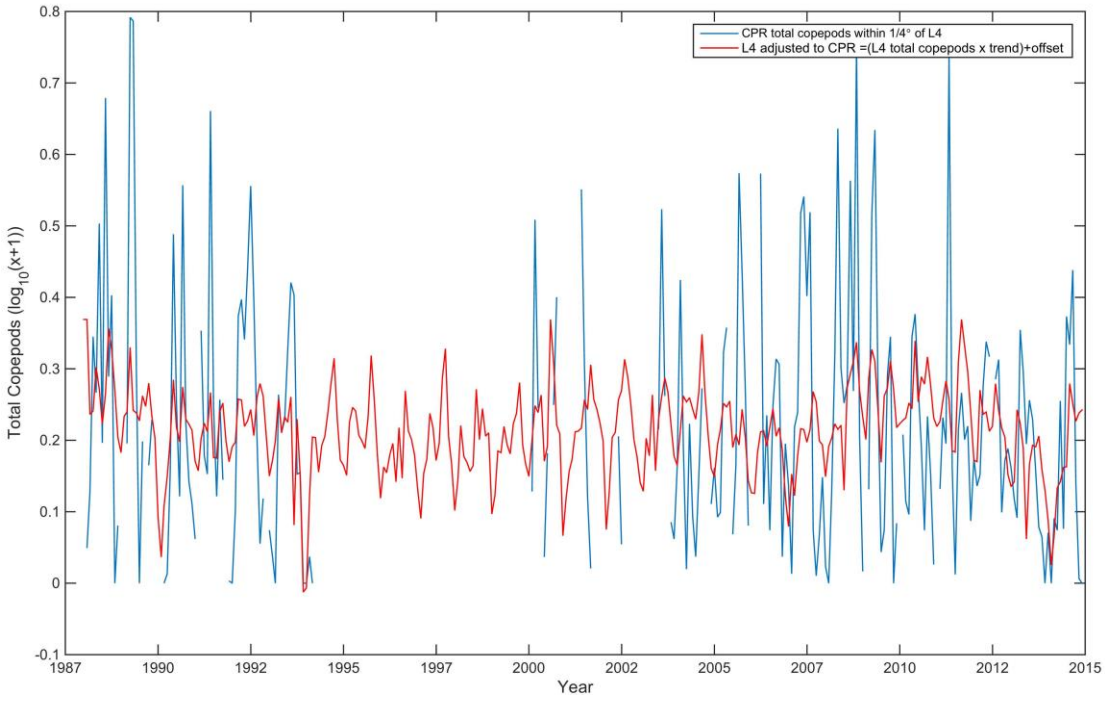

Figure 6. Monthly total copepod abundance for CPR samples within ¼ degrees of L4 = blue, and L4 total copepod abundance adjusted to CPR using the relationship between L4 and CPR samples within ¼ degrees = red.

Chl *a* and PCI did not show a significant relationship (pAC <0.01). This could be due to the categorizing of PCI values, which can be clearly seen for the CPR PCI within 1/8 and ¼ degrees of L4 (**Figure 7**). The strongest significance (pAC=0.016) was between CPR PCI values within 1 degree of L4.

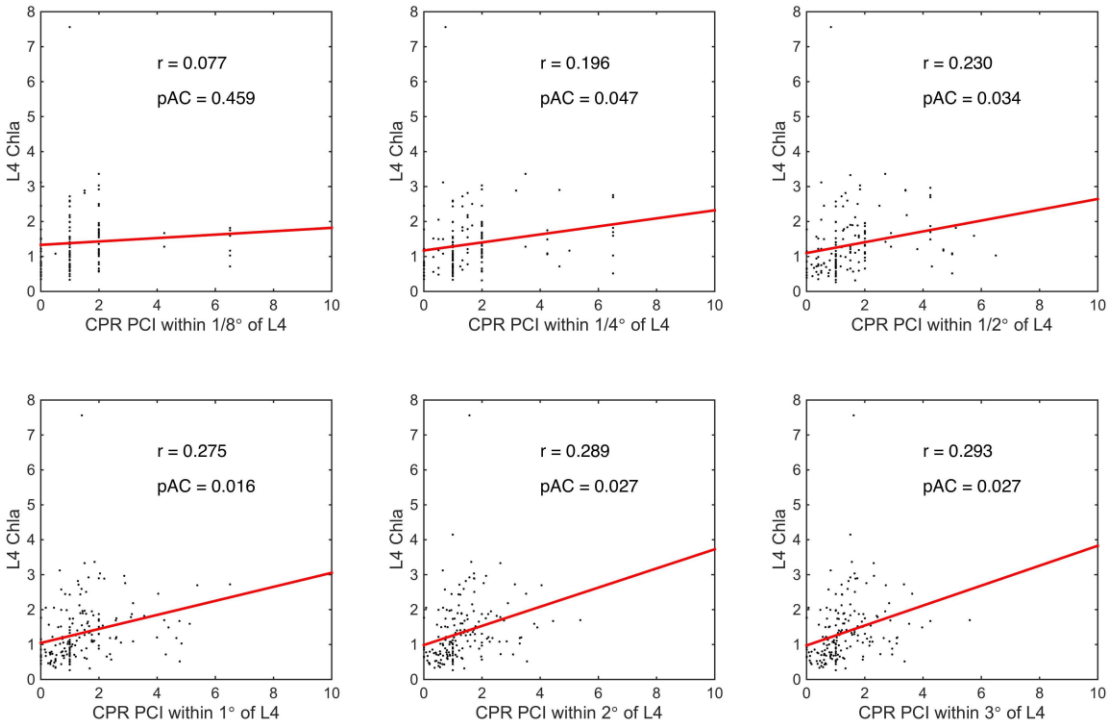

Figure 7: Relationships between chl *a* from L4 and PCI from the CPR dataset at different spatial scales. Pearson's correlation coefficient is displayed for each relationship as r, and the significance corrected for temporal autocorrelation as pAC.

4.3 Mean seasonal cycle

The mean seasonal cycle for total copepod abundance and total phytoplankton were calculated for each CPR spatial range (see **Figure 1**) and compared with that of L4. Mean seasonal cycles for total copepod abundance are shown in **Figure 8**. The L4 data show a double peak in abundance, with a larger peak following the spring bloom, during the late autumn months. The CPR data also show a double peak in abundance, however the larger of the two peaks occurs in the spring instead of the autumn. The spatial range that best matches the L4 mean seasonal cycle is the CPR data within ¼ degrees of L4.

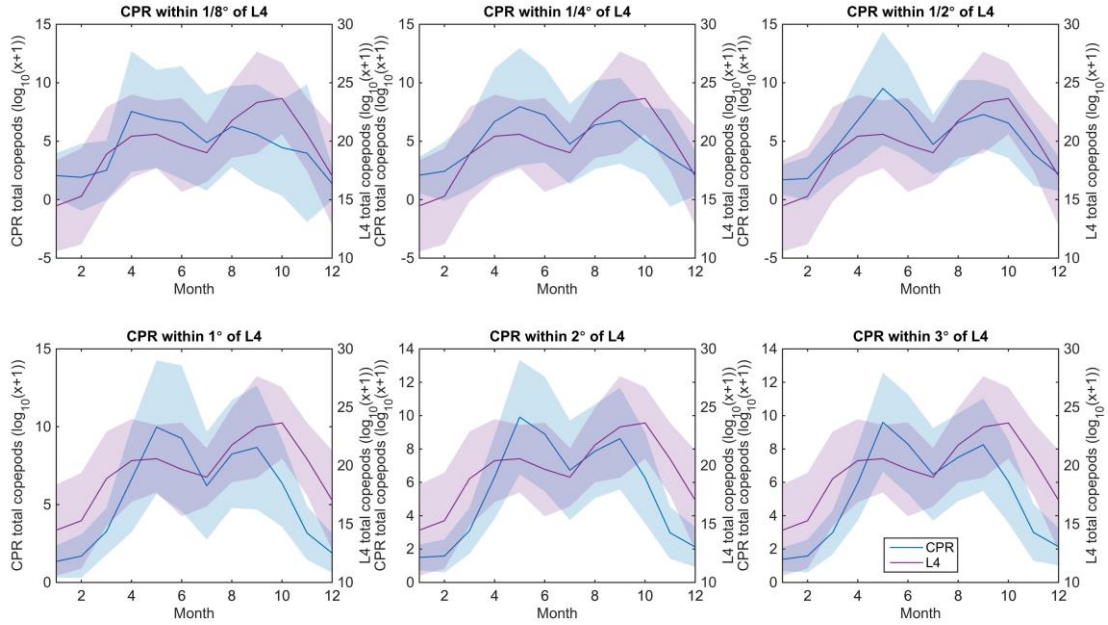

Figure 8: Mean monthly seasonal cycle of total copepod abundance. Purple = L4, blue = CPR samples at the different spatial ranges around L4 shown in **Figure 1.** The bounded area around the lines represents the standard deviation of the mean.

The mean chl *a* seasonal cycle at L4 shows a peak in February and another peak in chl *a* around June time, the lowest values of chl *a* are in the late autumn and winter months (see **Figure 9**). This double peak is most pronounced in the PCI CPR data that is closest to L4, within 1/8 degrees. Although this spatial range gave the lowest significance when analysing the monthly relationships as a time-series (**Figure 7**), when averaging the mean seasonal cycle it is most representative of the chl *a* at L4.

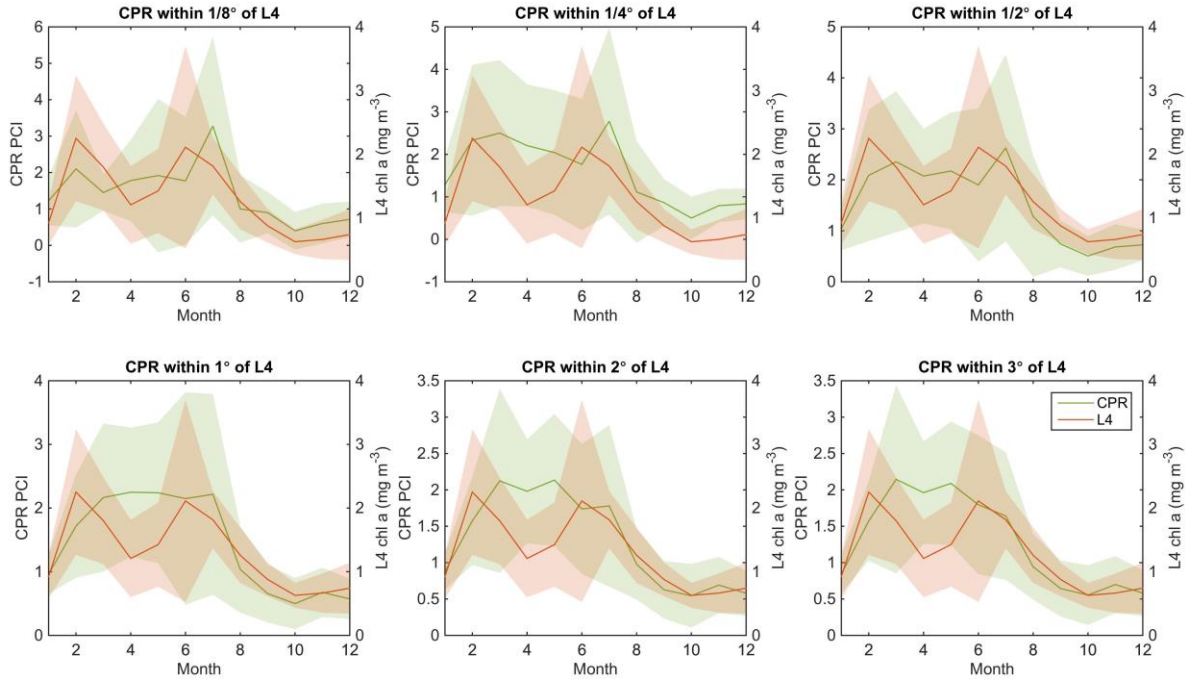

Figure 9. Mean monthly seasonal cycle of chl *a* from L4 and PCI from CPR data. Red = L4, green = CPR samples at the different spatial ranges around L4 shown in **Figure 1.** The bounded area around the lines represents the standard deviation of the mean.

4.4 Annual anomalies (PH2)

The annual anomalies for total copepod abundance were calculated by subtracting the mean seasonal cycle from the monthly data and averaging the monthly anomalies by year, this process is shown in **Figure 10** for the L4 dataset.

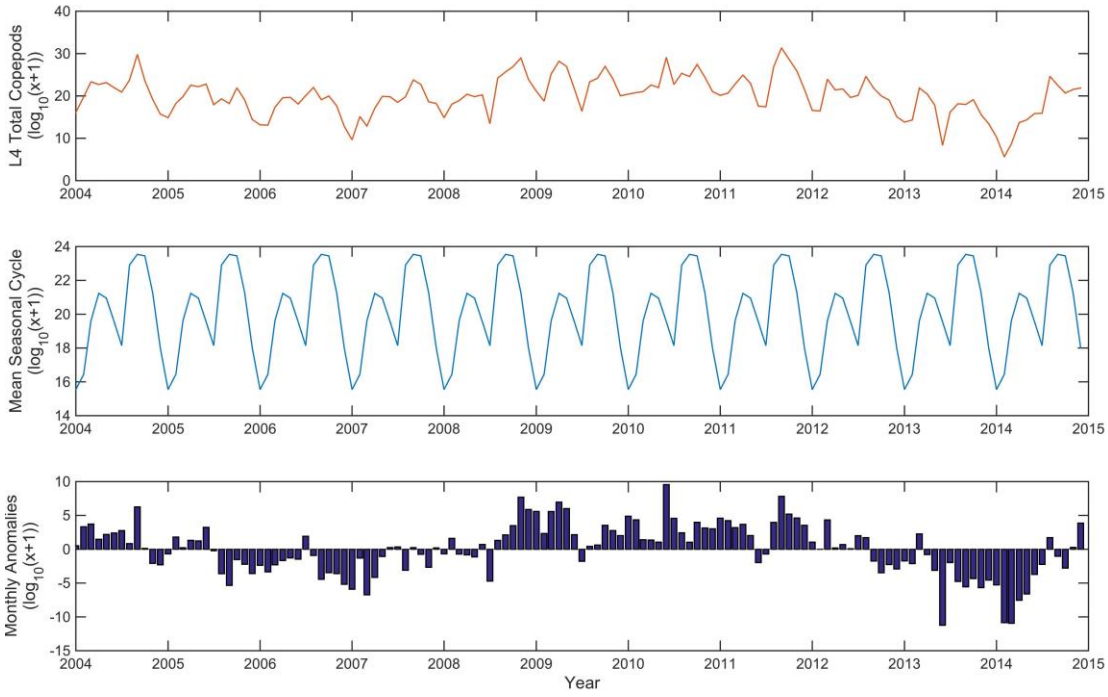

Figure 10: Top subplot = monthly total copepod abundance from L4. Middle subplot=Monthly mean seasonal cycle of total copepod abundance repeated for the time-series. Bottom subplot=Monthly anomalies, calculated by subtracting the mean seasonal cycle from the monthly time-series.

The annual anomalies of total copepod abundance for L4 and the CPR data surrounding L4 are shown in **Figure 11.** Although the magnitude is greater for L4 compared to the CPR data (this is also shown in **Figure 3**) most annual anomalies agree on the direction of change. 2004 is an exceptional year where L4 data suggests that copepod abundance has increased, whereas the CPR data suggests that it has decreased. The CPR data within a range of 1 degree from L4 have the most similar annual anomalies (**Figure 11**).

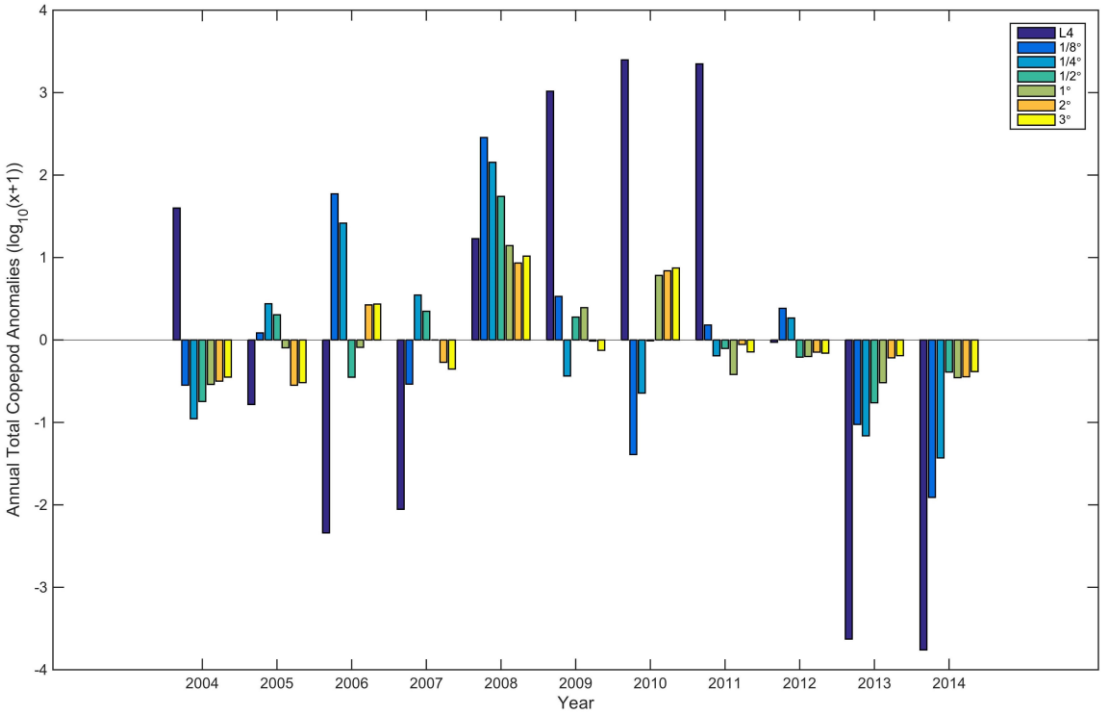

Figure 11: Annual anomalies in total copepod abundance for L4 and CPR data within the spatial ranges of L4 shown in **Figure 1** for the years 2004 to 2014.

The same process was also carried out for the comparison of annual anomalies for chl *a* from L4 and CPR PCI around L4 (**Figure 12**). 2008 and 2011 are years where the L4 data and CPR data do not agree on the anomalies, and the CPR data closest to L4 (within 1/8 degrees) gives the most similar results to the L4 chl *a* dataset.

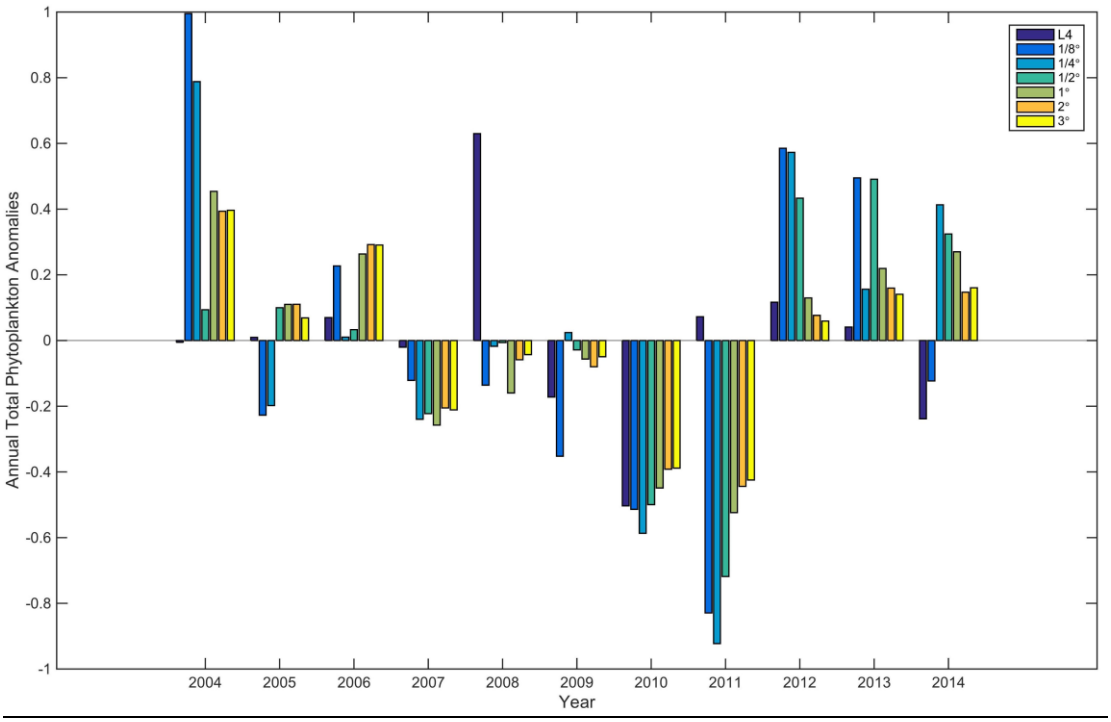

Figure 12: Annual anomalies in chl *a* for L4, and CPR PCI data within the spatial ranges of L4 shown in **Figure 1** for the years 2004 to 2014.

4.5 Annual Plankton Index (PH1)

As well as grouping the data into total copepods and using proxies for total phytoplankton, the data were also aggregated into lifeforms (see **Table 1**). These lifeforms were used to calculate the annual PI for the lifeform pairs described in **Table 1**. For the purpose of this report, two examples of these results are given, however the annual PI was calculated for each of the lifeform pairs available from both datasets and are provided within the Annex 1 of this report. **Figure 13** shows the annual PI for the lifeform pair small copepods and large copepods from 2004 to 2014. The CPR data surrounding L4 for this lifeform pairing agrees well during the reference period (2004 to 2008). The following years are less well matched, but the overall trend in the PI is similar between the different datasets.

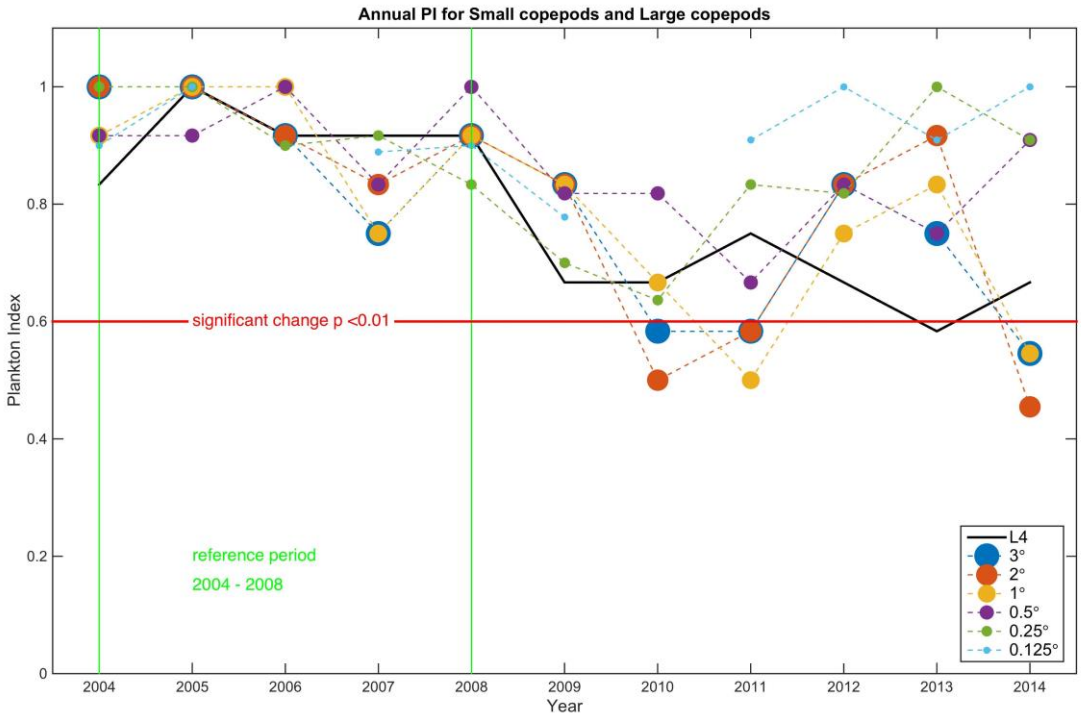

Figure 13: Annual PI values for the lifeform pair small coepods against large copepods for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change ($p<0.01$) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

Figure 14 shows the annual PI for the lifeform pairing between diatoms and dinoflagellates. The smallest radius around L4 (1/8 degrees) has gaps in the annual PI time-series, and therefore does not have a complete reference period, so should not be used for this analysis. The most well matched annual PI values to that of L4, are the CPR values within 1 degree radius of L4, with the exception of 2014 where 3 degrees and 0.25 degrees give a better match to L4. As is also the case with **Figure 13**, although specific years do not always match between the CPR spatial aggregations and L4, the general trend of the annual PI is similar between both datasets.

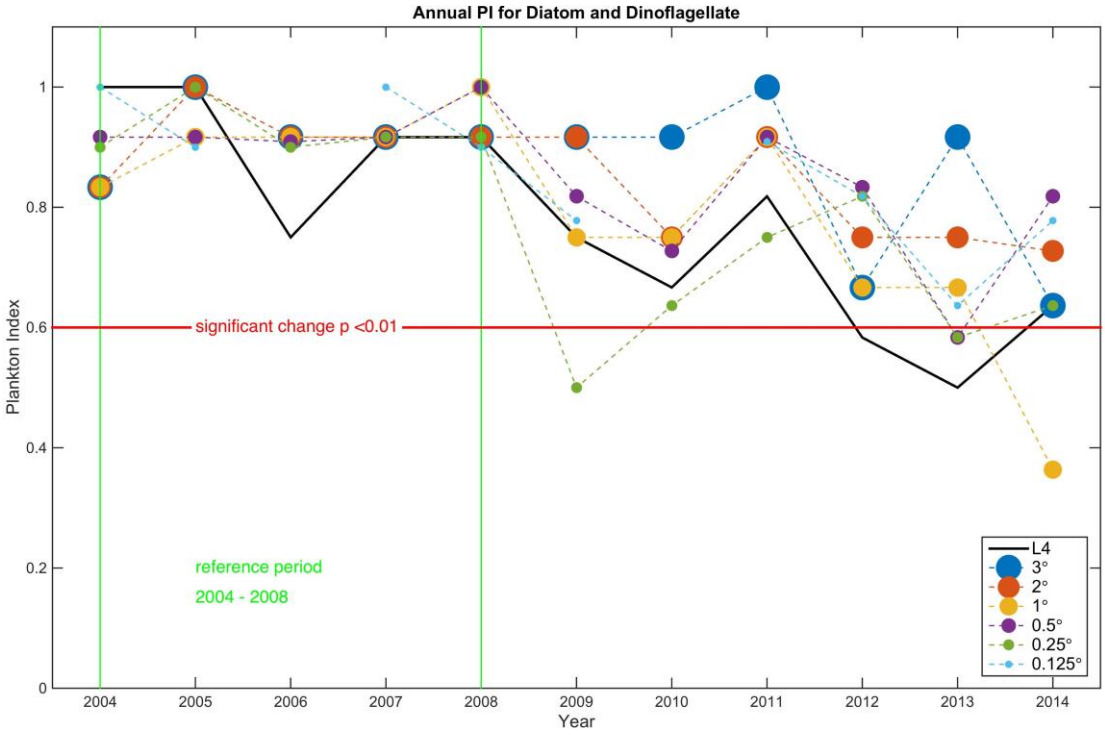

Figure 14: Annual PI values for the lifeform pair diatoms against dinoflagellates for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change ($p<0.01$) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

5 Discussion

Both sources of data investigated provide valuable and complimentary information; CPR data covers a wider spatial and temporal range, giving more regional long-term information, while fixed-point stations provide more detailed information on localised conditions. From the above analyses it is clear that plankton are patchy in their distribution, and their spatial variation limits the ability to merge differing datasets inter-annually, concluding that the use of 'conversion factors' becomes ambiguous. However, on mean seasonal and long-term time scales both datasets show strong similarities. From the analyses within this report it is apparent that there is a balance between having an adequate number of samples for aggregation within a close enough region around the fixed-point station to make a meaningful comparison.

Main conclusions:

This work based on a single case study, concludes that plankton are patchy in their distribution in space and time, and therefore the use of conversion factors is ambiguous. Long-term and seasonal trends can be compared and inferred using both sets of data, however fixed-point and transect data have differing complimentary information for time-series investigations.

Recommendations:

The report suggests the inclusion of both fixed-point and transect plankton data within assessments due to their complimentary nature. Caution should be taken when merging datasets and aggregating data as there is a balance between having enough data to represent a region as well as being able to pick up on key changes in plankton assemblages.

Gaps and further work:

As further development to this study the following recommendations are suggested:

- PH3 was not included in this analysis, as it was still under development. Once the biodiversity indicators used within PH3 have been defined, these should also be included within the spatial representivity analyses to provide a more holistic comparison between indicators and to investigate potential integration of the suite of indicators.
- Further investigation should be carried out to determine the optimum sample size to use with CPR data for all pelagic indicators (PH1, PH2, PH3). It is likely that this will vary between regions and indicators used, as physical dynamics play a strong role in the spatial variation of plankton. Therefore this shall have to be carried out on a site-by-site basis, however, a protocol for this determination of optimum sample size should be developed. This could be carried out by analysing the stability of the average at the lowest resolution using a boot-strapping technique.
- Ecohydrodynamic (EHD) areas (van Leeuwen *et al.,* 2015) should be used to test how representative samples from the same EHD are.

- A state-space version of a multivariate autoregressive model has been developed that accounts for observation error (Holmes *et al.*, 2010), whether this adaptation can be applied to the state-space approach used in PH1 should be investigated.
- If there is potential to adapt the plankton index state-space approach to account for observational error, perhaps a framework to incorporate spatial variation could also be incorporated in to the methodology for PH1.
- To more accurately define the spatial representivity of a fixed-point station, more information is required on the physical conditions surrounding the station and further analyses of individual taxa and their range in relation to those physical conditions is necessary.
- A further analysis should be carried out incorporating another more open-ocean station, E1. Both L4 and E1 employ the same sampling techniques for plankton, and could be used in conjunction with CPR data to provide more information on the influence of differing sampling techniques when comparing plankton indicator outputs.

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ANNEX 1

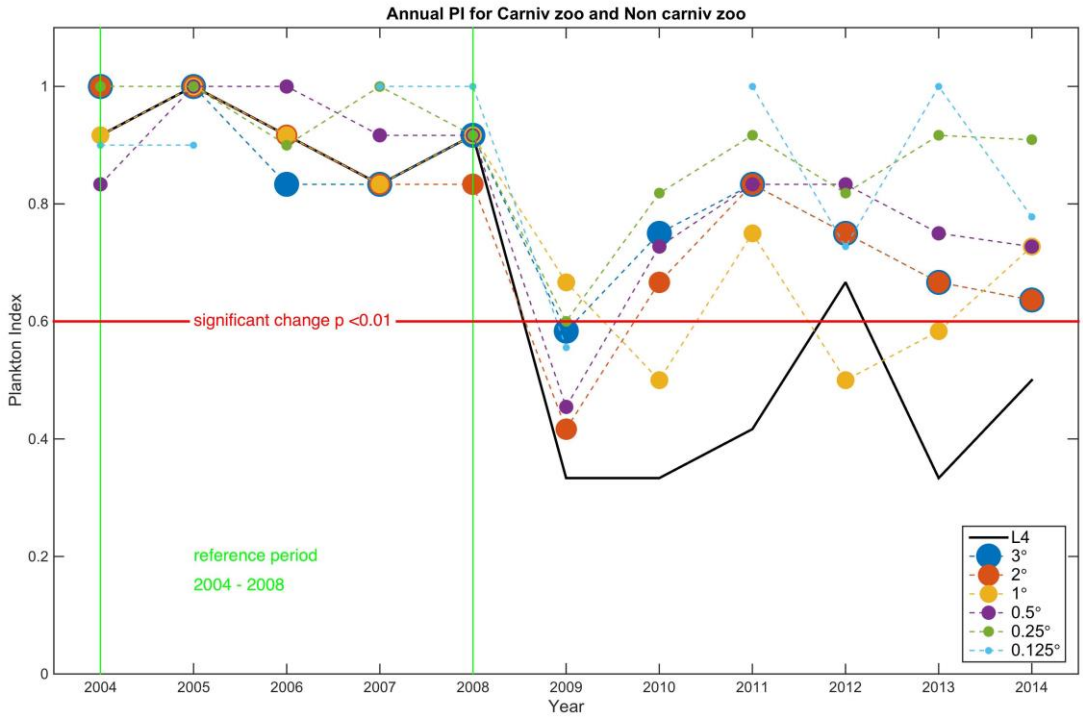

Figure A.1: Annual PI values for the lifeform pair carnivorous zooplankton and non-carnivorous zooplankton for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change ($p<0.01$) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

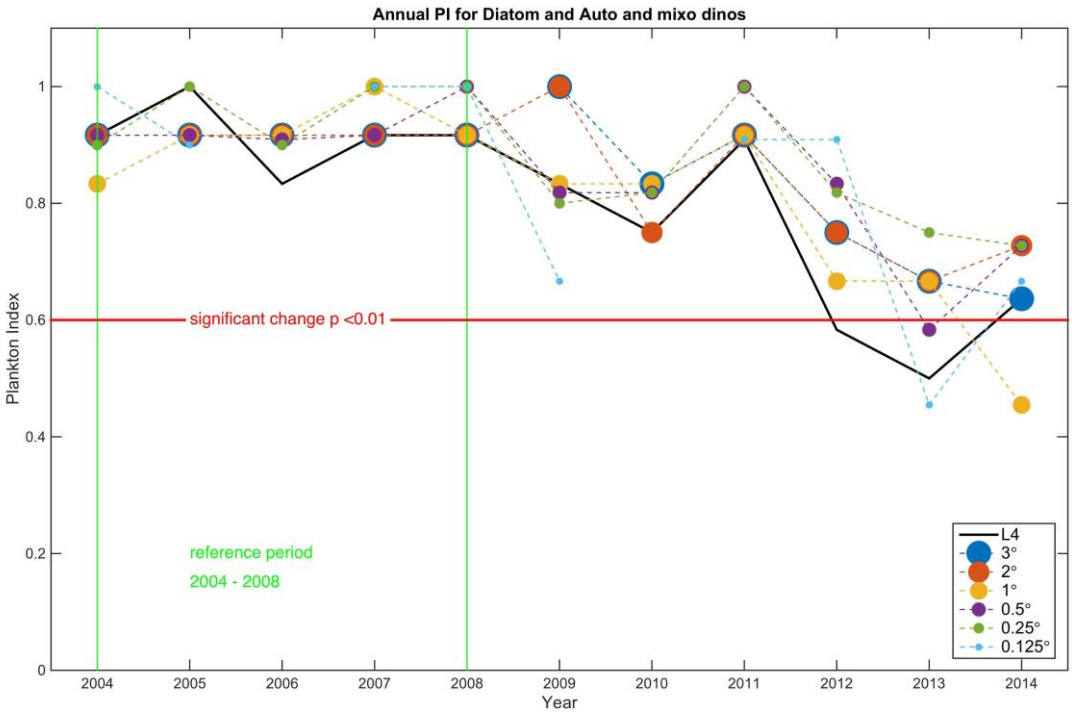

Figure A.2: Annual PI values for the lifeform pair diatoms and auto and mixo-trophic dinoflagellates for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change ($p<0.01$) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

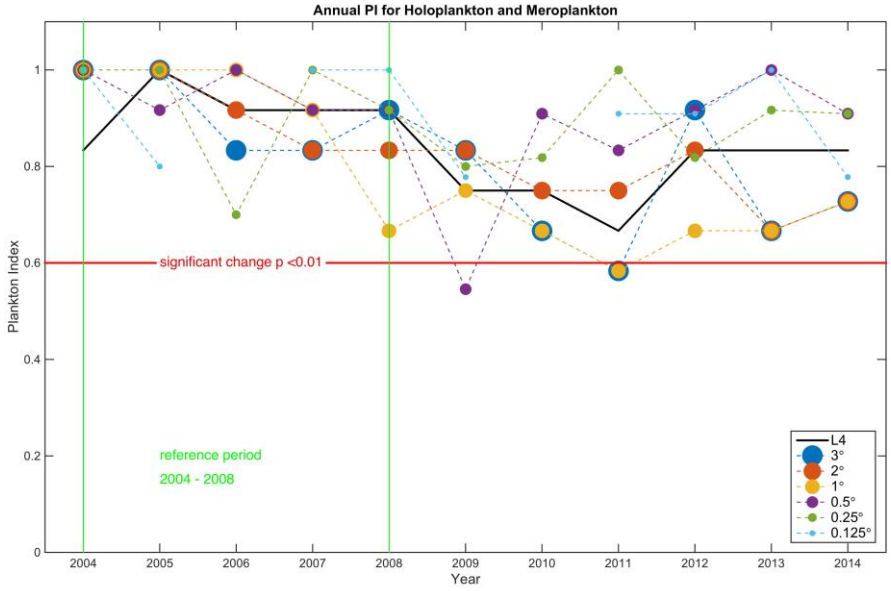

Figure A.3: Annual PI values for the lifeform pair holoplankton and meroplankton for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant

change (p<0.01) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

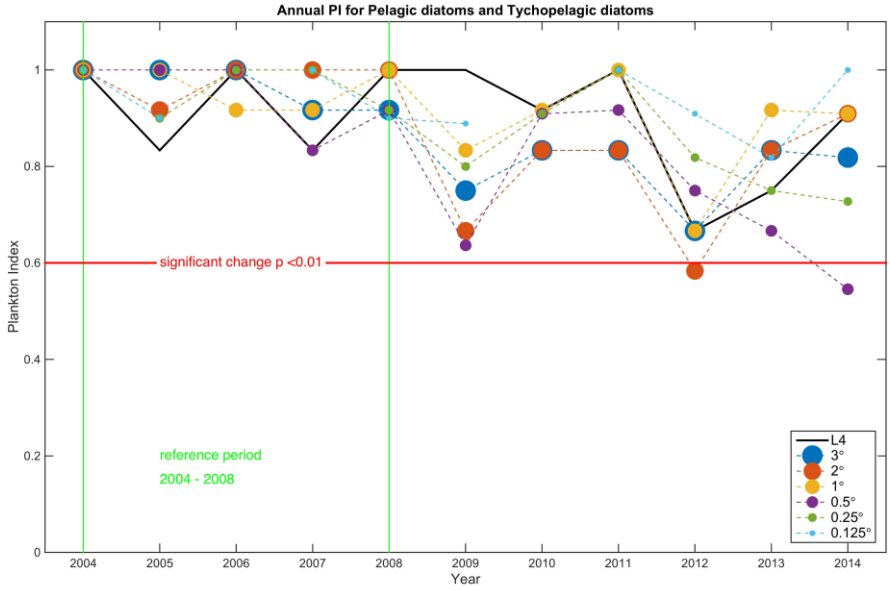

Figure A.4: Annual PI values for the lifeform pelagic diatoms and tychopelagic diatoms for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change (p<0.01) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

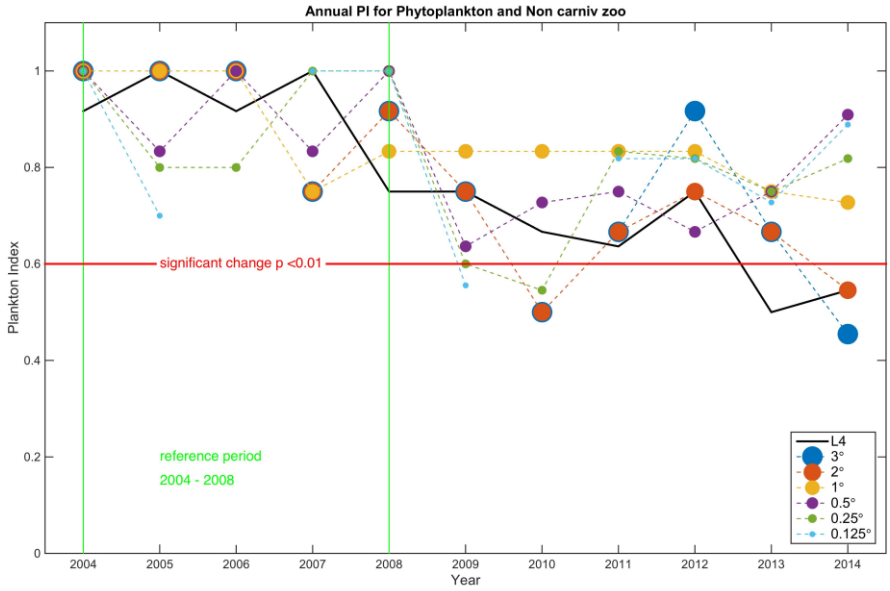

Figure A.5: Annual PI values for the lifeform pair phytoplankton and non-carnivorous zooplankton for L4 and CPR data within the spatial ranges shown in **Figure 1**. Black line = L4, dark blue circles = 3 degrees, red circles = 2 degrees, orange circles = 1 degree, purple circles = ½ degrees, green circles = ¼ degrees, cyan circles = 1/8 degrees. The reference period used for this analysis was 2004 to 2008, and falls between the green lines. A significant change ($p<0.01$) in the annual PI is characterised by a drop in the PI value to below 0.6, this threshold is represented as the red line.

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