BIOACID
Biological Impacts of Ocean ACIDification

Consortium 1: Pelagic ecosystems
WP 1.14: Adaptations to ocean acidification (OA) in mesozooplankton

Metabarcoding results from a long-term mesocosm experiment in the North Sea

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Introduction

Marine plankton is a very important component in the worlds ecosystem. Phytoplankton species are responsible for half of the world's photosynthesis and remove approximately 100 million tons of carbon dioxide from the Earth per day. Zooplankton furthermore play a key role in marine food webs as they transfer energy captured by phytoplankton to higher trophic levels. The ongoing acidification process of the oceans may have consequences for this important marine biota. Possible impacts could for example be changes in community composition since there are probably more or less CO_2 sensitive species. Furthermore there could be changes in the succession and abundance of species for example due to a slower development and reproduction.

In our study we used **metabarcoding** to investigate the impacts of OA for different reasons. First of all there is a potentially high diversity of cryptic species as well as larval stages which are not captured by morphological investigations. Secondly metabarcoding datasets not only include information about the occurrence of different species but also about genetic variation within those species. **The application of genetic techniques to obtain a very high level of taxonomic resolution allowed us to uncover community changes under CO_2 stress.**

Mesocosm Experiment

Sampling
From 17 m depth to the top of each mesocosmos
Mesoplankton (>200 µm): Apstein net with 55 µm mesh size, fixed in 90% EtOH
Microplankton (<200 µm, >0.45 µm): 500 ml water from an integrative water sampler filtered onto a 0.45 µm nylon filter, fixed in 90% EtOH

Metabarcoding sample selection

1,5,9 ambient CO_2 cosms / 2,4,6 high CO_2 cosms / 10 fjord

Time points: t17, t41, t65, t97

Marker regions: 18S, cox1, cox2

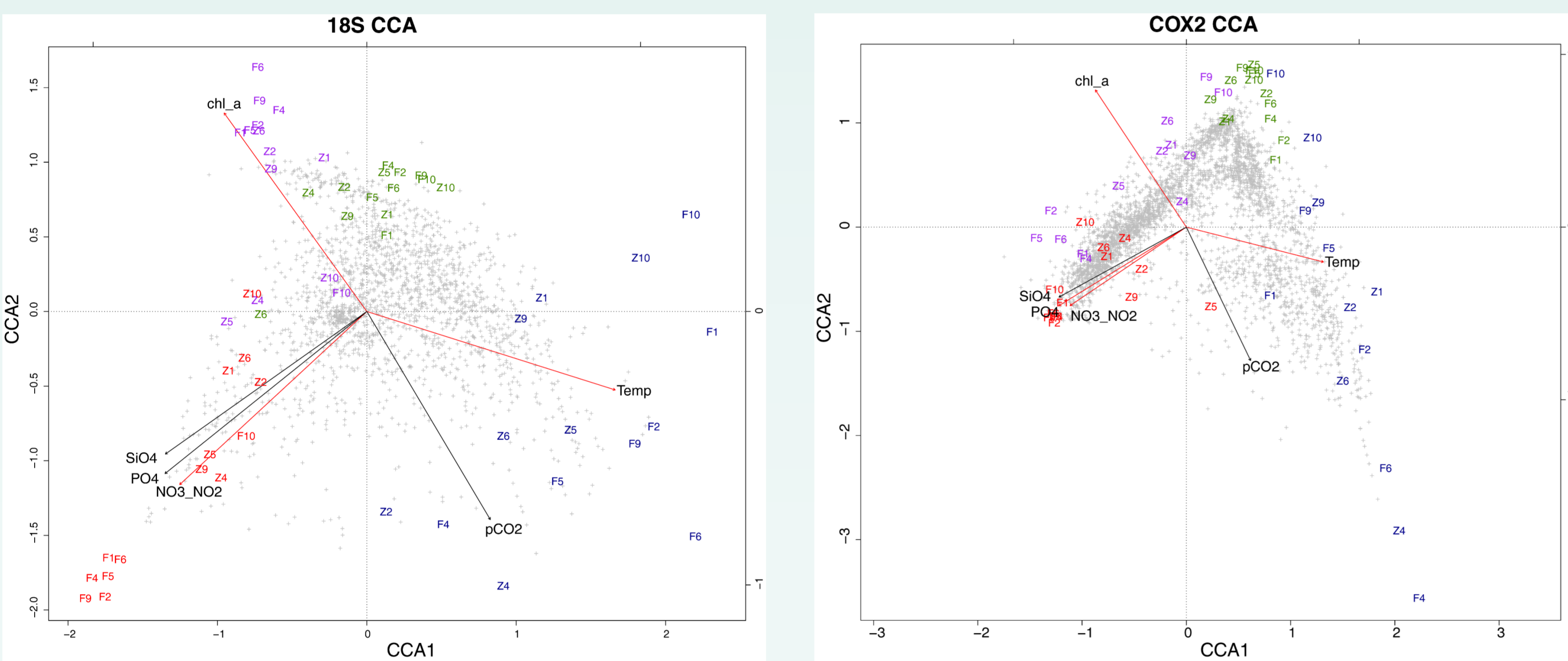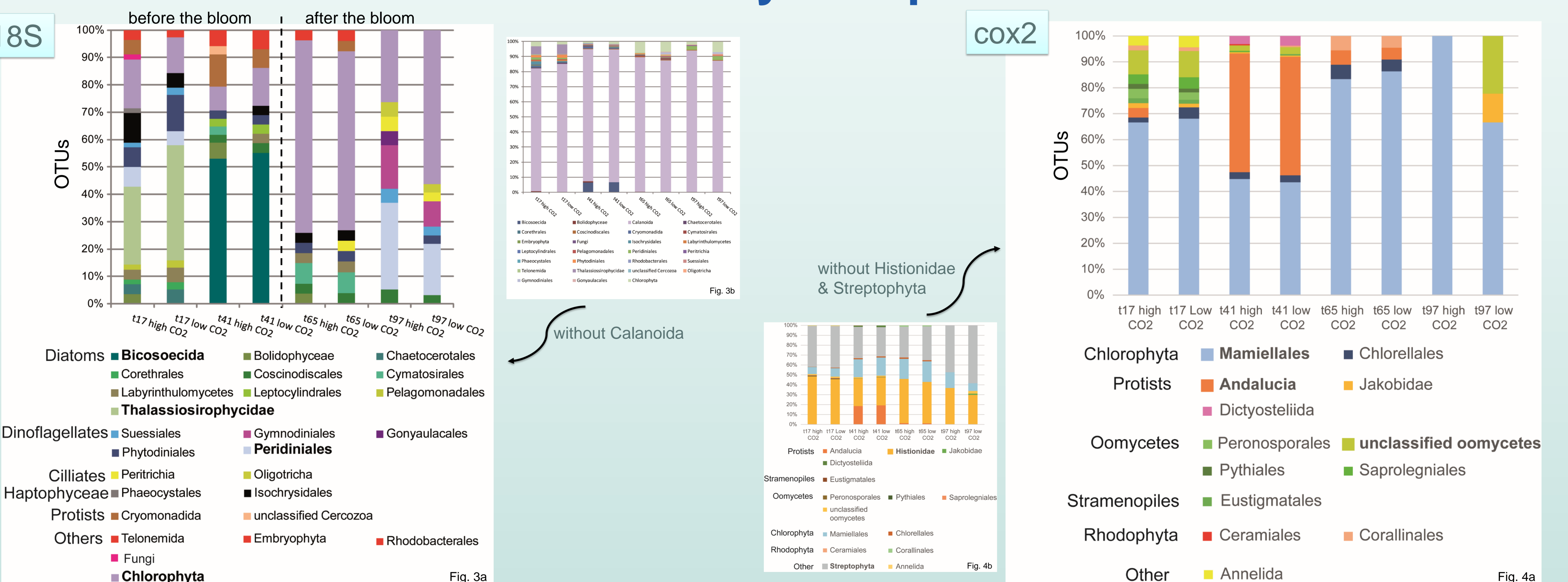## Results: Environmental Parameters and CO_2

Fig. 1 & 2 CCA based on the 18S (1) and cox2 (2) data. OTUs (+); arrows represent the environmental variables (significant ones are marked in red); numbers represent the mesocosms and the attached character defines wether it is a filter (F) or net sample (Z); time points are separated by different colors t17 (red), t41 (purple), t65 (green), t97 (blue).

We conduced CCAs (based on Chi-squared distances) using binary OTU abundance matrices of the 18S (Fig. 1) and cox2 (Fig. 2) data to check whether the OTU composition can might be explained by environmental variables which were measured during the mesocosmos experiment (significant variables are marked in red).
At the first time point (red values) the OTU composition is mainly depending on nutrients. With raising temperature and light intensity an algae bloom started to develop within the mesocosms which coursed a change in the OTU composition (purple values). After the bloom phase most of the nutrients were consumed and most of the OTU variation is explained by the reduction of nutrients (green values). Towards the end of the experiment with still rising temperature the OTU composition of the mesocosms started to differ from each other (blue values).

The micro- and mesoplankton community show sparse differences in their OTU composition between the CO_2 treatments but a clear change over the time due to changes in nutrient supply and temperature.

Results: Mesocosms Community Composition

18S

Fig. 3a

Fig. 3b

without Calanoida

Diatoms: **Bicosoecida**, Bolidophyceae, Chaetocerotales, Corethrales, Coscinodiscales, Cymatosirales, Labyrinthulomycetes, Leptocylindrales, Pelagomonadales, **Thalassiosirophycidae**
Dinoflagellates: Suessiales, Gymnodiniales, Gonyaulacales, Phytodiniales, **Peridiniales**
Ciliates: Peritrichia, Oligotricha
Haptophyceae: Phaeocystales, Isochrysidales
Protists: Cryomonadida, unclassified Cercozoa
Others: Telonemida, Embryophyta, Rhodobacterales, Fungi, **Chlorophyta**

cox2

Fig. 4a

without Histionidae & Streptophyta

Fig. 4b

Chlorophyta: **Mamiellales**, Chlorellales
Protists: **Andalucia**, Jakobidae, Dictyosteliida
Oomycetes: Peronosporales, **unclassified oomycetes**, Pythiales, Saprolegniales
Stramenopiles: Eustigmatales
Rhodophyta: Ceramiales, Corallinales
Other: Annelida

In a BLAST search 80% of the 18S OTUs assigned to the group Calanoida (Fig 3b). Beside that we found mainly different Diatoms, Chlorophytes, Dinoflagellates and Ciliates (Fig 3a). **Different CO_2 treatments do not remarkably differ in taxa composition.** The main shift in taxon composition occurred during the algae bloom. After the bloom we found a significant higher percentage of chlorophytes and a significant lower percentage of diatoms (t-test: $p<0.05$). Additionally, at the last time point dinoflagellate and ciliate diversity raised.

Based on the cox2 data we found a high percentage of OTUs which assigned to the family Histionidae and the group Streptophyta (Fig. 4a). Furthermore we mainly found Chlorophyta, Rhodophyta and Oomycetes (Fig. 4b). **The composition of taxa based on the cox2 OTUs show again sparse differences between the CO_2 treatments but a clear change over time.**

Future perspective
➤ Identification of candidate OTUs which differ among CO_2 treatments
➤ Cryptic diversity within species complexes
➤ Phylogenetic mapping

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