

Climate Change Effects on Plankton Communities in the Baltic Sea: A Mesocosm Experiment

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Degree Thesis

Thesis for NOVIA UAS – BSc Sustainable Coastal Management
Sustainable Coastal Management Program

Ekenäs, 2023

DEGREE THESIS

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Degree Programme and place of study: BSc Sustainable Coastal Management, Ekenäs

Specialisation:

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Title: Climate Change Effects on Plankton Communities in the Baltic Sea: A Mesocosm Experiment

Date: 6.6.2023 Number of pages: 42 Appendices:

Abstract

The study focussed on analysing potential impacts of climate change on plankton communities in the Baltic Sea. Phytoplankton being the primary producers in the marine trophic system, and zooplankton being the trophic link to higher levels of the food web, have critical functional roles. Shifts in their communities could potentially have significant impacts in the ecosystem and even ecosystem services. For this study, a five-week indoor mesocosm experiment was set up to simulate projected warming and changing salinities in the Baltic Sea. Freshwater and seawater were collected and mixed in 600 L containers and were subjected to two temperature scenarios (18°C and 21°C) at different salinity levels to represent climate change effects. My results show that temperature had a significant relationship with fluorescence (proxy for phytoplankton biomass) and with zooplankton species richness. Specifically, diatom groups such as Navicula sp. and Thalassiosira sp. as well as cyanobacteria Anabaena sp. (now known as Dolichospermum sp.), and Merismopedia sp. were found to have significant relationship with salinity. In addition, the presence of Vorticella sp. was observed in all warm treatments. Overall, the experiment concludes that plankton community shifts are driven by abiotic factors such as temperature and salinity in line with climate change effects. For the Baltic Sea, results showed that Keratella sp. have high adaptability and can thrive wider range of salinity and temperature scenarios while Anabaena sp. can benefit from the slow freshening of the Baltic Sea.

Language: English

Key Words: climate change, zooplankton, phytoplankton, plankton communities, plankton ecology

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1 Introduction

Among the numerous global threats in our marine ecosystems including pollution, eutrophication, and biodiversity loss, climate change remains to be one of the major issues that persists with its potential damaging environmental impacts (Andersson, et.al., 2015). Although the impacts of present changes in the environment were studied to vary geographically (Doney, et.al., 2012), studies on climate change by the BACC II Author team (2015) have suggested the potential significant impacts of climate change in the Baltic Sea.

Changing environments brought about by climate change can drive population-level shifts and altered trophic pathways, due to changing ecological interactions, changing dispersal patterns, and physiological tolerance (Doney, et.al., 2012). Furthermore, to understand better the impacts of climate change effects, projection model projects such as the CMIP focussing on different future scenarios based on policy-targeted goals, are currently used to assess potential large-scale climate variations in the Baltic Sea (Meier, et.al, 2022).

1.1 Historical trends for temperature and salinity in the Baltic Sea

The Baltic Sea has experienced a slow freshening of its surface waters since the 1960s of approximately 0.01-0.02%/yr (0.1-0.2 ppt/yr) (Kankaanpää, et.al., 2023). Furthermore, decrease in salinity may be attributed to increased freshwater inputs over the past six decades which is expected to continue with the potential climate change effects. It has been a dominant feature of the Baltic Sea that during the Anthropocene period, the diminishing trend on surface and near bottom salinity is expected to cause ecological disturbances including marine food webs (Kankaanpää, et.al, 2023). As environmental variations can affect species, habitats, and ecosystems, climate change remains to be a clear driver of how the Baltic Sea environment will be formed in the succeeding decades. According to recent studies, the present average surface salinity of the Baltic Sea is seven grams per kilogram of water (SYKE, 2020).

Interestingly, while the surface-level temperature remained to be consistent over the past decades, the trend on near-bottom layer of the Baltic Sea has been observed to increase

by 0.75-2.9°C as documented from the 1960s up until to 2021, where results strengthen the notion of increasing climate change impacts (Kankaanpää, et. al.,2023).

1.2 Projected trends in the Baltic Sea

Future climate change assessments were presented in Meier's et.al. 2022 report, where it is expected to affect numerous phenological processes in relation to the Baltic Sea. An example is the shift of phytoplankton spring and summer blooms brought about warming (Visser and Both, 2005). Surface air temperature, precipitation, cryosphere, sea level, water temperature and salinity, and marine biosphere were included in the variables observed during the study. Future modelling assessments showed possible scenarios in the next decades relating to climate change with their corresponding impacts in the Baltic Sea.

According to HELCOM (2021), sea surface temperature is anticipated to continuously rise throughout the Baltic Sea, along with freshwater influx. In addition, the Bothnian Sea, Gulf of Finland, and Gulf of Riga are expected to experience significant ice cover decline due to warming and higher precipitation caused by climate change, leading to possible increased stratification, and change in nutrient dynamics in the trophic system.

Projected estimates from Meier (et al., 2022), were presented regarding climate change, indicating a potential sea level rise, from years 2081-2100, where the global mean sea level (GMSL) expected to range between 0.40m-0.63m, primarily driven by thermal expansion and melting of glaciers. In addition to their study, projected ocean warming is expected to increase within a range of 1°C to 3°C, and 0.5°C to 1.5°C at a 1 km depth in the next century. Furthermore, their reported projected salinity in the Baltic Sea shows a decreasing trend, as freshening is driven by the melting of ice and intensified water cycles. While some of these projections on climate change are presented with high confidence, many factors are recognised and the actual impacts have yet to determine.

Specifically in the northern Baltic Sea region including Bothnian Bay, Bothnian Sea, and Gulf of Finland, projected changes in water temperatures are greatest in the summer season for the two former regions and spring season for the latter (Meier, 2015). In addition, summer

sea surface temperature (SST) can potentially rise to 2-4 °C in the southern and northern Baltic Sea, respectively (Meier, 2015). Lastly, projected salinity variations by multi model ensemble simulations at the sea surface is relatively small in both the northern Baltic Sea (Meier, 2015).

1.3 Phytoplankton and their ecological importance

Phytoplankton are microscopic algae or bacteria, which are the largest photosynthetic group that form the foundation of the aquatic food web (Suikkanen et.al., 2013). Consisting of around 1,700 species in the Baltic Sea (Ojaveer, et.al., 2010), they are crucial in the ecosystem functioning.

In the context of environmental processes, phytoplankton have a significant role in the global carbon cycle. Being photosynthetic organisms, most species take up CO₂ during the process while some are considered mixotrophs where they depend on other sources for nutrition (Engel, 2020). Globally, phytoplankton are a key player in the biological carbon pump and can fix forty percent of the global carbon annually (Falkowski, 1994).

Given the critical functional role of phytoplankton in the aquatic ecosystem, impacts on phytoplankton are monitored and reported in line with potential impacts of climate change (BACC II author team, 2015).

1.4 Zooplankton and their ecological importance

Zooplankton are microscopic animals that are found in water bodies such as rivers, lakes, and seas. Their role in the marine ecosystem is considered pivotal to the food web, and nutrient cycling (Dam and Baumann, 2018). This is brought about by different processes such as feeding, excretion, and respiration that contribute to their functional role in the community (Steinberg and Landry, 2017). For instance, feeding on primary producers can influence phytoplankton communities and their production (Dam and Baumann, 2018). According to Calbet (2001), mesozooplankton consume about 12% of the global marine primary production. The feeding process bridges the primary producers to higher trophic levels, such as invertebrates, and fishes, marine mammals (Dam and Baumann (2018); Steinberg and Landry, (2017)). This process is associated with transfer of nutrients and

energy from one trophic level to the next level. In terms of nutrient cycling, wastes through the form of excretion and respiration from zooplankton contribute to the amount of nitrogen, phosphorus, and carbon for recycling and energy transport (Steinberg and Landry, 2017).

With the clear function of zooplankton in the ecosystem, they are usually monitored for their influence on algal densities, and nutrient cycling (Dam and Baumann, 2018) to anticipate any possible cascading events in the ecosystem.

1.5 Impacts of climate change on phytoplankton and zooplankton

According to the study of Suikkanen et.al (2013), recent observations show the changing species composition and abundance of phytoplankton in the Baltic Sea. Aside from climate change effects, their study suggested that eutrophication and other top-down pressures play a role in influencing the changes. It was further suggested that species composition changed in spring bloom where diatoms:dinoflagellate ratio decreased. Along with this, increased harmful phytoplankton blooms are expected.

On the other hand, climate change can have direct impacts on zooplankton through warming, acidification, and deoxygenation, further affecting the physiology, behaviour, life history traits and reproduction to adapt to the environmental changes (Dam and Baumann, 2018). Temperature adaptation has been observed in the copepod *Eurytemora affinis* (Karlsson and Winder, 2020); their results showed that species had less adaptation potential than the same species that have already been thriving in warming temperatures. This supports the results from previous studies showing that zooplankton from the tropical regions can have higher adaptability than the polar and subpolar regions due to the limited genetic capacity to adapt to increasing temperatures (Somero, 2012).

In a recent study, zooplankton community evenness and composition were found to reorganize in response to changing salinity (Hall and Lewandowska, 2022). Their outdoor mesocosm experiment showed shifting dominance of groups exposed in higher salinity,

contrary to low salinity scenarios where there were no shifts observed. Rotifers were found to dominate low salinity areas which are mostly present in freshwater environments. As suggested, their abundance will likely increase when the projected trend on Baltic Sea freshening takes place.

1.6 Phytoplankton Taxa

There are over 1,700 species of phytoplankton in the Baltic Sea ranging from the smallest picoplankton, nanoplankton, and macroplankton (Ojaveer, et.al, 2010). Each of them belongs to groups which are categorized to have distinct characteristics and functions in the ecosystem. In terms of function, phytoplankton can be categorized based on their trophic type: autotrophs, heterotrophs, and mixotrophs. In addition, they can also be categorized based on size. According to Brotas et.al (2022), there are three classifications used to group the sizes of the phytoplankton: picoplankton (diameter <2 μ m), nanoplankton (diameter from 2 to 20 μ m) and microplankton (diameter >20 μ m) (Sieburth et al., 1978).

For this research, I focused on identifying and counting taxa listed below, taking into account the common visible species to provide a general picture on their community changes under climate anticipated variations.

1.6.1 Diatoma

Diatoms are single-celled algae, which are identified through their silica-formed cell walls for DNA replication (Wu, et.al., 2022; Sexton & Lomas, 2018). They are a highly diverse group common in freshwater, marine, and moist terrestrial ecosystems, which contribute 40% to the global primary production (Wu, et.al., 2022, Sarthou et al. 2005). They are considered the most important phytoplankton group being key players in the carbon cycle (Sarthou et al. 2005). Morphologically, species in this group have diverse characteristics and vary in shape (single cells or chains) and size, varying from micrometers to millimeters.

6

Diatoms are grouped into three classes: Coscinodiscophyceae, Bacillariophyceae, and

Mediophyceae (Medlin, and Kaczmarska, 2019).

In the context of climate change effects, a decrease in salinity was suggested to increase

diatom diversity; however, decreasing their productivity (Virta and Teittinen, 2022). And

during warming, spring bloom consisting of diatoms and dinoflagellates starts earlier,

followed by an earlier and prolonged summer bloom dominated by cyanobacteria

(Viitasalo, et.al., 2015).

For the purpose of this research, I have focused on Navicula spp., Nitzschia sp., Synedra

spp., Fragillaria sp., Thalassiosira sp. and Tabellaria sp.

Class: Bacillariophyceae

The Bacillariophyceae are mostly photosynthetic pennate diatoms, but few are categorized

as mixotrophs and heterotrophs, contributing to the carbon cycle (Kociolek et.al, 2015). In

this taxon, is a group of pennate diatoms that have a raphe system which is a morphological

slit visibly found on one or both valves of a diatom. This allows the diatoms to move at

different speeds and distances. Major groups under this order are Eunotiales,

Achnanthales, Naviculales, Bacillariales, Rhopalodiales, and Surirellales (Kociolek et.al,

2015). Generally, raphid diatoms are found in different habitats like freshwater, marine,

tidal mud flats, and areas where have high range of salinity, pollution, temperature and

substrate types (Kociolek et.al., 2015).

Class: Coscinodiscophyceae

Coscinodiscophyceae are described to contain radially-symmetric, centric diatoms.

Thalassiosira sp. with a diameter 20-100μm (Olenina et.al, 2006) are primarily found in

marine waters. Other genera include Coscinodiscus, Rhizosolenia, and Melosira.

Araphid Diatoms: Fragilariophyceae and Tabellariales

This colony-forming subclass of diatoms are classified separately from the centric and raphid diatoms. However, they are determined to be closely related based on cladistic analysis (Williams, 1990). I have included the genera *Fragilaria* sp. and *Tabellaria* sp. for this research as part of the diatom group.



Figure 1 . (A) *Nitzschia* sp., (B) *Nitzschia* sp. (C) *Fragilaria* sp., (D) *Navicula* sp. observed under the Leica TV 8 inverted microscope.

1.6.2 Dinophyta

Dinoflagellates are microplankton with a diameter ranging from 40-85 μ m according to the Biovolumes and Size-classes of Phytoplankton in the Baltic Sea Handbook (2006). With their varying sizes and shapes, dinoflagellates are most commonly known to bloom and become the source of red tide that affect fish and marine animal mortality. However, their ecological function remains important as they are autotrophs involved in photosynthesis, carbon fixation, and energy cycles (Carty, and Parrow, 2014).

Although there have been studies regarding this group showing that they are favoured warmer sea conditions (Spilling, et.al., 2018), where species have been found to dominate during warm winters, other studies have suggested that it is not applicable to large-scale climate patterns as they may also depend on local environments, and local community structure (Klais, et.al., 2016).

For this research, I focused on the genus Ceratium sp. (diameter of 40-85µm).

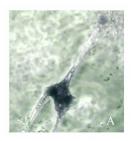


Figure 2. (A) *Ceratium* sp.observed under the Leica TV 38 microscope at 10x magnification at 12.6x objective

1.6.3 Chlorophyta

This class is composed of non-motile and colonial green algae commonly described as little green balls (Shubert, 2003) that can easily be misidentified with different species. However, morphological features allow the differentiation between species. Chlorophytes are widely distributed and abundant in freshwater environments during summer (Shubert, 2003). Different species dominate depending on the season like for *Dictyosphaerium* sp. (diameter 3-7,5µm), possibly dominating shallow nutrient-rich water lakes in summer (Shubert, 2003).

Along with other groups, chlorophytes were reported to have increased in the summer season in northern Baltic region as an effect of climate change where sea temperature rose and salinity decreased from 1979-2008 (Viitasalo et.al., 2015).

1.6.4 Cyanophyta

Cyanobacteria are considered to be the oldest organisms on Earth and one of the most important photosynthetic group of microorganisms because of their contribution and functional role in both freshwater and marine ecosystems (Vincent, 2009; Komárek and Johansen, 2014). They consist of chlorophyll-a and several phycobilin-protein complexes to facilitate photosynthesis (Komárek and Johansen, 2014). Usually, they are blue-green in colour that can be observed from the satellite view of the oceans during a season of bloom. In terms of functional role, this diverse and abundant group is critical in the carbon fixation as they are CO₂-dependent, and provided that they are generally abundant, the group

contributes greatly in primary production, nitrogen fixation, production of toxic compounds and their symbiotic roles in the ecosystem (Komárek, and Johansen, 2014). With the high diversity and abundance of the cyanobacteria, several species can be found worldwide, each thriving in different environmental requirements and depend other ecological factors. Their importance increases during blooms and eutrophication resulting to substantial biomass and toxin production. *Microcystis, Nodularia,* and *Trichodesmium* are some of the common species found in brackish coastal waters (Komárek and Johansen, 2014).

Warming of the Baltic Sea, along with the decrease in salinity and the increase of nutrients, such as nitrogen and phosphorus from winter melting, were the primary factors in increased summer biomass in the northern Baltic proper, including the Gulf of Finland (Viitasalo et.al, 2015).

For this research, I identified the following genera: *Anabaena* sp. (now known as *Dolichospermum* sp.) (diameter 3.5-12μm), *Oscillatoria* sp., *Merismopedia* sp. (diameter 0.5-6μm), and *Gomphosphaeria* sp.

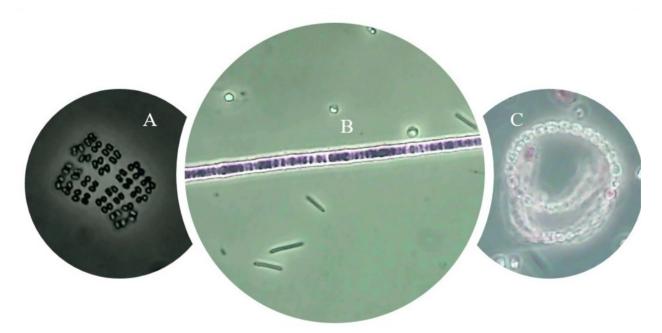


Figure 3. (A) *Merismopedia* sp. (B) *Oscillatoria* sp.(C) *Anabaena* sp. Viewed from the Leica TV 38 inverted microscope

1.7 Zooplankton taxa

In the brackish environment of the Baltic Sea, the dominant zooplankton groups include copepods, rotifers, and cladocerans, which are part of the mesozooplankton group (Telesh et. al., 2015). Out of forty species, 10 to 12 are considered dominant groups in the Baltic Sea as they thrive in different salinity ranges (Telesh et. al., 2015). They are classified based on sizes: nano zooplankton (2-20µm), microzooplankton (20-200µum), mesozooplankton (0.2-20mm), and microzooplankton (>20mm) (Hernroth, 1981). With climate change effects over the past decades, shifts in species composition were observed and different patterns were noted for the abundance ratio between cladocerans and copepods; however, for the Gulf of Finland, the trend was seen to clearly decrease (Telesh, et.al, 2015). The research focused on the following groups:

1.7.1 Copepoda

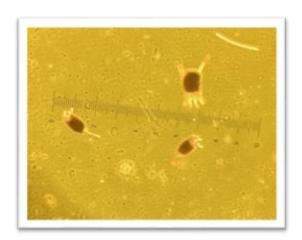


Copepods are microscopic planktonic crustaceans that are between 0.5 and 5 mm in size and that make up 70-90% of mesozooplankton abundance (Turner, 2004; Steinberg and Landry (2017). There are around 63 taxa of free-living copepods in the open Baltic Sea (Telesh et. al., 2009). Classified into three subgroups: Calanoida, Cyclopoida, and Harpacticoida, they have different shapes (elongated, fusiform, cylindrical), sizes, ecology and feeding patterns (Telesh et. al., 2009). In terms of their role in the food web, they feed on algae, smaller invertebrates, and rotifers (Brandl, 2005). Previous studies such as Hall and Lewandowska (2022) show that copepods can tolerate changes in salinity differently and a restructuring in the community may occur in higher salinity scenarios.

1.7.2 Cladocera

Cladocera include crustaceans ranging from 0.2 to 3 mm, and they are primarily common in freshwater bodies (Telesh et. al., 2009). Particularly in the Baltic Sea, there are 37 identified taxa listed and the genera *Bosmina* spp. was found to be most common (Telesh et. al., 2009). In addition, most cladocerans are able to tolerate a wide range of temperatures and low oxygen concentrations of less than one part per million (Telesh et. al., 2009). Examples of cladocerans are *Daphnia*, *Bosmina*, *Diaphanosoma*, *Holopedium*, *Leptodora* for freshwater and *Podon*, *Pleopsis* and *Evadne* for the Baltic Sea. Sea surface warming was suggested to have a positive relationship in terms of growth for cladocerans, especially during summer (Viitasalo et al. 1995).

1.7.3 Rotifera



Rotifers are microscopic invertebrates mostly found in freshwater bodies, but live also in both brackish and marine environments, usually comprising the majority of the biomass (Telesh et. al, 2009). Although many species of rotifers are sessile, most are free swimming, preying on microalgae (12-18µm), bacteria, and detritus (Telesh et.al, 2015). The majority of the rotifer species are solitary and some form colonies. They are able to reproduce fast, contributing as food source to higher trophic levels, such as other invertebrates and fish larvae (Telesh, et.al, 2009). There are a total of 83 Rotifera taxa identified in the open Baltic Sea based from the Zooplankton of the Open Baltic Sea: Extended Atlas (2009).

Similar to cladocerans, rotifers were found to be positively affected by warming of the sea surface (Viitasalo et al. 1995), whereas a study in the northern Baltic proper, increased rotifer abundance was observed with decreasing salinity (Suikkanen et. al., 2013).

2 Research Question

Since zooplankton are the trophic link from the phytoplankton to the invertebrates, linking the complex food-web altogether, their importance in the ecosystem is deemed critical. Therefore, studying plankton community shifts with changing abiotic conditions brought about climate change is essential to understand possible impacts not only in the food web but its implications to the ecosystem services it provides.

Focusing at the community level will also provide information on how different species thrive and interact together- which species are affected negatively and which ones can adapt to the changes and the physiological stresses from the change in temperature and salinity.

My study aims to answer:

- 1. Are phytoplankton groups affected by the temperature and salinity changes related to climate change?
- 2. How will zooplankton communities shift with changes in both temperature and salinity brought about by climate change?

Further Research Necessary

The threat of climate change in the Baltic Sea environment includes the elevation of sea temperature, decreasing salinity, resulting to increased stratification, and eutrophication. With this, further research is necessary to understand how these climate-driven variations can impact plankton communities in the Baltic Sea. Furthermore, my study aims to focus specifically on the southwest coastal region of Finland, understanding phytoplankton and zooplankton community shifts with the threat of climate change impacts.

3 Methods

3.1 Mesocosm set-up

To test the research question, I participated in an indoor mesocosm setup during the summer season, for five weeks (35 days) from September until October 2022 to study the effects of changes in temperature and salinity on plankton communities in Tvärminne Zoological Station in Hanko, Finland. This project was under the lead supervision of Aleksandra Lewandowska and Clio Hall.

The experiment consisted of two different temperature scenarios of 18.0 °C and 21.0°C, and six salinity scenarios of 3.0 (control), 4.5, 6.0, 7.5, 9.0, and 10.5 PSU. The scenarios were based on both ambient and predicted warming and salinity change from Meier et al. (2012) that were adjusted using the MONICOAST monitoring buoy data to depict local conditions. The water used in the experiment was a mix of 50-50 proportion of freshwater and sea water of 300L each. Freshwater was collected from Gennarbyviken reservoir having 0 psu, whereas seawater was collected in the bay in front of Tvärminne Zoological Station to represent the marine environment of the Baltic Sea (5.5 psu). Assessments were conducted to ensure that the communities survived and that they were similar in each setup. To adjust from the initial salinity to different scenarios, sea salt (Instant Ocean) was added in batches and mixed carefully using a Secchi disk to achieve the desired salinity. Two temperaturecontrolled climate chambers were used to maintain the set room temperatures throughout the experiment. Six 600L containers with specifications of 200 um thick, with a dimension of 1.25m x 0.5m x 1m were installed in each room to represent the six levels of salinity for this project. LED lamps (customized LED spot AquaMedic controlled by Proflux units, GHL) were positioned above each treatment setup to mimic natural light conditions.

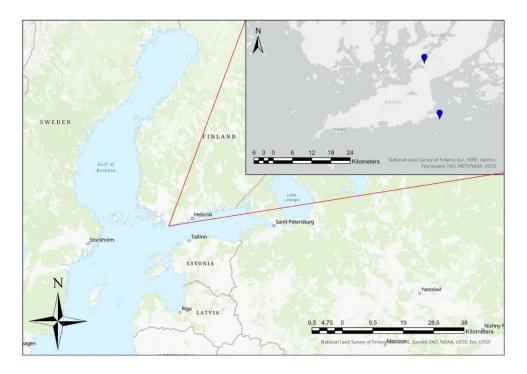


Figure 4. Map of water collection sites and Tvärminne Zoological Station (Produced using ARC Pro, 2023)

The overall focus of the mesocosm experiment is to analyse the effects of temperature and salinity change on plankton food webs in pelagic ecosystems. However, for my research, I focused on four specific scenarios: 3.0, 4.5, and 6.0 psu in warm temperature, and 6.0 psu in the cold temperature. Narrowing down to these four scenarios was the focus of my research on climate change effects which was based on the current average salinity of Baltic Sea and the general projections of warming and freshening of the Baltic Sea from Meier et. al. (2022).

Water sampling was conducted once a week and temperature, salinity, and chl-a were measured every two days from DOY 251 to 279. Each container was mixed using a Secchi disk before transferring 5L water to 10-L plastic containers. The containers were gently mixed before taking the subsamples. Falcon tubes were used to collect 50ml for each sample and acid Lugol's solution was added to maintain the structure of the plankton. For my research, a total of 20 samples were collected from the five-week experiment and were stored in the fridge for microscopy.

3.2 Microscopy

Microscopy work was done according to the HELCOM Guidelines on Phytoplankton Monitoring Handbook (2021), using the Utermöhl method (Edler and Elbrächter, 2010). A sediment chamber and a 50ml cuvette were used to settle the samples for 24 hours. Cells were counted, using Leica TV 38 inverted microscope. Cell identification and counting was based on Kasviplanktonopas (1986), Helcom Checklist of Baltic Sea Phytoplankton Species (Hällfors, 2004), and Helcom Biovolumes and size-classes of phytoplankton in the Baltic Sea (Olenina et.al., 2006). Other online sources (i.e Nordic Microalgae) were also used to crosscheck and confirm the groups being identified.

Identifying and Quantifying Zooplankton and Phytoplankton

Zooplankton and selected phytoplankton were identified at the genus level and counted using the whole chamber method at low magnification of 10x objective (HELCOM, 2021). This was applied to larger taxa namely *Copepoda, Cladecorans, Rotifers*, and specific phytoplankton taxa namely *Tabellaria* sp., *Fragilaria* sp., and *Ceratium* sp.

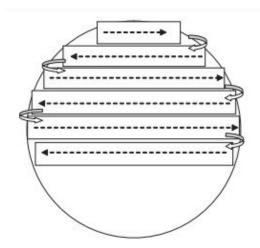


Figure 5. Identification and counting using the whole chamber method from Edler & Elbrächter 2010, applied to larger sized taxa. (Photo from UNESCO, 2010)

Intermediate sized taxa were counted using a modified diameter transect method from the Utermöhl method (Edler and Elbrächter, 2010; UNESCO, 2010). Selected phytoplankton taxa were also identified at the genus level and counted per cell estimate using the transect method, modified to randomly select five views at 40x magnification for each of the nine sections (See figure 6), with the aim to cover as much area as possible. A total of 45 field of views per sample were performed for this method. The following phytoplankton taxa counted for this method were: *Anabaena* sp., *Nitzschia* sp., *Navicula* sp., *Synedra* sp.,

Dictyosphaerum sp., Merismopedia sp., Gomphosphaeria sp. and Thalassiosira sp. The selected taxa for identification represent different phytoplankton groups, and dominant genera were included which was is supposed to be enough to provide a general picture of the community changes in the different scenarios. Unfortunately, smaller taxa were not identified and counted for this research due to my ability and time constraint.

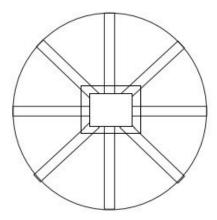


Figure 6. Transect method used for the identification and counting intermediate sized taxa

The formula of the phytoplankton density coefficient was obtained from HELCOM Guidelines for Monitoring Phytoplankton Species Composition, Abundance and Biomass Handbook (2021). The formula for the coefficient C is shown below:

Formula: $C(dm^{-3}) = A*1000 / (a^2*V)$

A = cross-section area of the top cylinder of the combined sedimentation chamber

a² = total counted area

 $V = volume (cm^3)$ of sedimented aliquot

The result from this formula, which is the coefficient, was multiplied with the number of cells estimated for each genera found in each sample to reflect the cell density. To compare the densities of the species, bar graphs were plotted to present the total count for each week. For this research, it is important to emphasize that the identification of the abovementioned taxa are according to the best of my knowledge with the understanding that they may be not 100% accurate.

Species richness and abundance

Apart from identifying and counting the species and units, species richness was determined by calculating the number of species in each sample, with abundance for zooplankton accounting for the total count of all individuals. To analyse the data, Anova test, T-test were used to compare species richness and abundance and determine whether they have significant difference.

Statistical analyses

To further analyse the relationship among factors with the groups, a regression analysis was used using MS Excel. Some graphs were presented using MS Excel while Origin Pro 2022 was used for plotting fluorescence and hydrology data.

4 Results

4.1 Hydrography

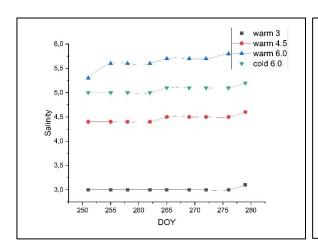
Temperature for all the warm treatments had an increasing trend throughout the experiment with maximum temperature at 21.2 °C at DOY 279. The water temperature had small variations as shown in Figure 7. The cold treatment on the other hand peaked at 17.3 °C with small changes throughout the experiment.

In terms of salinity, 3 and 4 psu treatments were close to the target salinity levels, however, for the 6.0C treatment, the salinity was closer to 5 (Figure 7).

Table 1. Salinity, water Temperature (°C) measurements

DOY	3.0W		4.5W		6.0W		6.0C	
	Salnity	Temp °C						
249	3	18.5	3	18.5	3	18.5	2.5	16.4
250	2.9	19.6	4.4	19.6	5.5	19.7	5.2	17.2
251	3	19.8	4.4	19.8	5.3	19.7	5	16.8
255	3	20.7	4.4	20.8	5.6	20.8	5	17.1
258	3	21.1	4.4	21.2	5.6	21.2	5	17.3
262	3	20.9	4.4	20.9	5.6	20.9	5	17.2
265	3	20.7	4.5	21	5.7	21	5.1	17.3
269	3	20.9	4.5	21	5.7	21	5.1	17

272	3	20.8	4.5	20.8	5.7	20.8	5.1	17.1
276	3	20.9	4.5	21	5.8	21	5.1	17.1
279	3.1	21.2	4.6	21.2	5.8	21.2	5.2	17.2



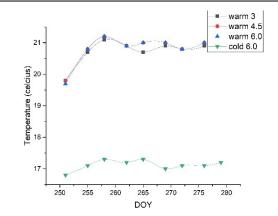


Figure 7. Salinity and temperature measurements for the four scenarios treatments (Raw data from Lewandowska and Hall, 2022)

4.2 Identified taxa

For this research, the following zooplankton and phytoplankton at the genus level have been identified and counted:

Zooplankton	Phytoplankton	Class	Trophic type
Acartia sp.		Copepoda	Heterotrophic
Cyclops sp.		Copepoda	Heterotrophic
Eurytemora sp.		Copepoda	Heterotrophic
Bosmina sp.		Cladocera	Heterotrophic
Daphnia sp.		Cladocera	Heterotrophic
Keratella sp.		Rotifera	Heterotrophic
Vorticella sp.		Oligohymenophorea	Heterotrophic
	Fragilaria sp.	Fragilariophyceae	Autotrophic
	Tabellaria sp.	Tabellariales	Autotrophic
	Navicula sp.	Bacillophyta	Autotrophic
	Nitzschia sp.	Bacillophyta	Autotrophic
	Synedra sp.	Bacillophyta	Autotrophic

Ceratium sp.	Dinophyta	Autotrophic,
		Mixotrophic,
		Heterotrophic
Anabaena sp. /	Cyanophyta	Autotrophic
Dolichospermum sp.		
Merismopedia sp.	Cyanophyta	Autotrophic
Gomphosphaeria sp.	Cyanophyta	Autotrophic
Thalassiosira sp.	Coscinodiscophyceae	Autotrophic
Dictyosphaerium sp.	Chlorophyta	Autotrophic

4.3 Phytoplankton

4.3.1 Salinity 6.0 at Ambient Temperature (18°C)

Phytoplankton community changed throughout the experiment as there was a significant decrease in abundance, particularly for *Tabellaria* sp., *Fragillaria* sp. and cyanobacteria *Gomphosphaeria* sp. As weeks passed, out of the 11 identified species in the first week, diversity sharply declined leaving one species left, *Synedra* sp., in the last week of the experiment.

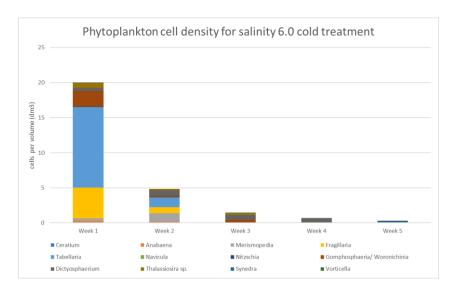


Figure 8. Phytoplankton composition for five weeks at 6.0 salinity and cold temperature

4.3.2 Salinity 6.0 at Warm Temperature (21°C)

Phytoplankton community for this scenario showed a steady decline from week 1 to week 5. Unlike the other treatment, the dominant species in this community at the beginning were *Tabellaria* sp., cyanobacterium *Gomphosphaeria* sp., and *Merismopedia* sp. Synedra sp. also remained at the end of the experiment.

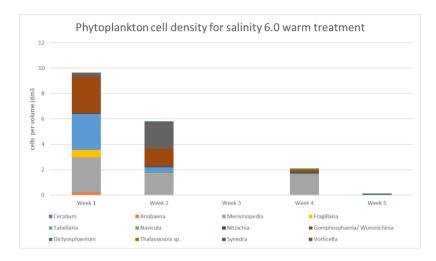


Figure 9. Phytoplankton composition for five weeks at 6.0 salinity and warm temperature

4.3.3 Salinity 4.5 at Warm Temperature (21°C)

For this scenario, the phytoplankton observations showed high numbers of *Fragillaria* sp. which declined in counts the following weeks. On the other hand, I found an increase in *Dictyosphaerium sp.*, *Gomphosphaeria sp.*, and *Tabellaria* sp. in the following week. Overall, a drop in the total cell density was observed in the succeeding weeks which may have contributed to the increase of zooplankton populations for week 3 (Figure 14). For this scenario, *Dictyosphaerium sp.* dominated the groups observed in the final week.

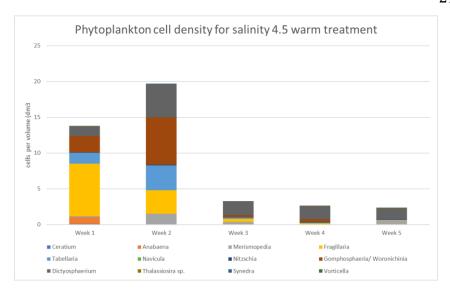


Figure 10. Phytoplankton composition for five weeks at 4.5 salinity and warm temperature

4.3.4 Salinity 3.0 at Warm Temperature (21°C)

The trend for this scenario is generally like the other treatments where a steady decline was observed from week 1 onwards. However, it was observed that after *Tabellaria* sp. dominated the first week of the experiment, it significantly declined in numbers after. This was followed by the other species, but interestingly, *Thalassiosira* sp. emerged to have increased in number and retained its presence until week 5. It is also interesting to find that cyanobacterium *Anabaena sp.*, increased in this treatment, dominating the community at the end of the experiment. Lastly, *Vorticella sp.* attached to other organisms in the set-up, were found to be highest in week 4 with 34 counts, but the population decreased a week later.

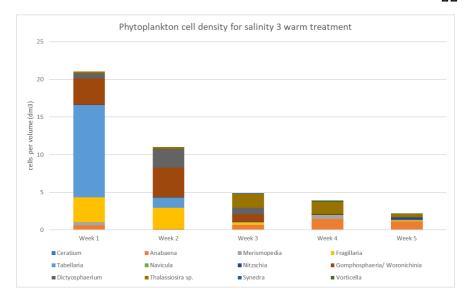


Figure 11. Phytoplankton composition for five weeks at 3.0 salinity and warm temperature

4.4 Zooplankton

4.4.1 Salinity 6.0 at Ambient Temperature (18°C)

The 6.0 salinity at ambient temperature scenario showed that the composition of zooplankton entirely changed from *Eurytemora sp.* and *Daphnia sp.* to *Keratella sp.* dominated community The cladoceran was not found during the following four weeks as rotifers started to appear and dominate the community. Some nauplii appeared in week 3 but did not remain in the succeeding weeks with rotifers completely dominating the community for this treatment.

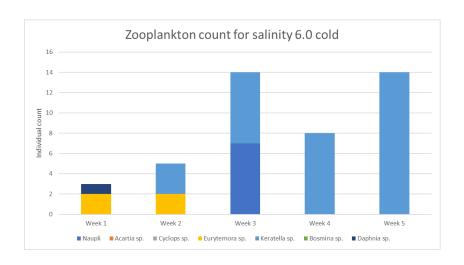


Figure 12. Zooplankton composition for 5 weeks at 6.0 salinity and cold temperature

4.4.2 Salinity 6.0 at Warm Temperature (21°C)

For this scenario, it was interesting to observe that even if the salinity of this scenario was the same as with the cold treatment, the result came out differently. Results show that the composition of the zooplankton was more diverse, dominated by adult copepods and nauplii. This treatment also showed population growth particularly for the nauplii at the end of the week. At the end of the experiment, the community was composed of copepod nauplii, *Eurytemora sp., Keratella* sp. and *Bosmina* sp. which are a mix of fresh water and marine species.

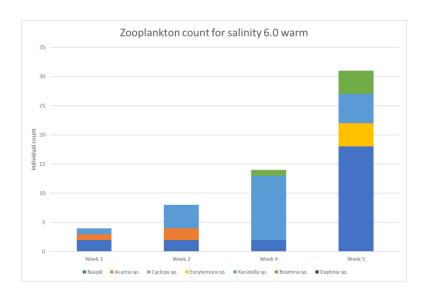


Figure 13. Zooplankton composition for 5 weeks at 6.0 salinity and warm temperature

4.4.3 Salinity 4.5 at Warm Temperature (21°C)

Results showed a composition of freshwater species *Keratella* sp., and *Bosmina* sp. in the first week. However, a significant increase of *Keratella* sp. was observed in the third week along with the presence of cyclopoid copepod *Cyclops* sp., and nauplii. However, a significant drop was observed in the *Keratella* sp. numbers along with the sudden presence of *Vorticella sp.* during week 4. Finally, the zooplankton community composition resulted

in increased numbers of nauplii, *Keratella* sp., and a reappearance of *Bosmina sp.* and Copepoda *Acartia sp.*, and *Eurytemora sp.* In terms of species richness, this scenario had the highest diversity in the last week of the experiment.

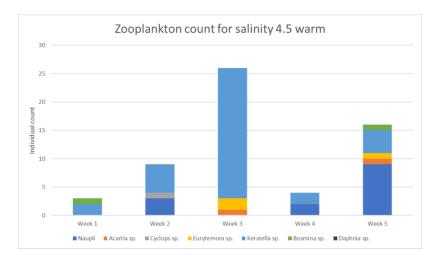


Figure 14. Zooplankton composition for 5 weeks at 4.5 salinity and warm temperature

4.4.4 Salinity 3.0 at Warm Temperature (21°C)

Salinity 3.0 at warm temperature scenario resulted in an overall dominance and presence of the freshwater species, *Keratella* sp. Every second week, the presence of copepods such as *Cyclops* sp., *Eurytemora* sp. and nauplii were observed. The growth of the community peaked on the third week when nauplii had appeared. At the end of the experiment, rotifers were found to outnumber the *Eurytemora* sp. that were observed.

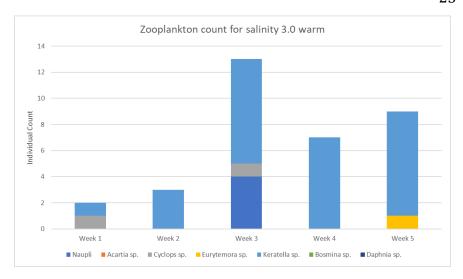


Figure 15. Zooplankton composition for 5 weeks at 3.0 salinity and warm temperature

4.5 Comparing Different Salinity and Temperature Scenarios

For the phytoplankton observations, a general steady and decreasing trend was observed for all samples from the first week to the fifth week. The dominant species *Tabellaria sp.* and *Fragillaria sp.*, as well as the larger-sized taxa such as *Ceratium sp.* sharply declined after the first two weeks and was replaced by other species such as *Dictyosphaerium sp.*, *Thalassiosira sp.*, *Merismopedia sp.* and *Gomphosphaeria* sp.. The composition of phytoplankton in the last few weeks varied where some cyanobacteria taxa, particularly *Anabaena* sp., dominated species in the low salinity treatment (3.0) in the last week of the experiment.

Other observations include the abrupt presence and disappearance of *Vorticella* sp. in all the warm treatment samples during weeks 3 and 4.

On the other hand, community composition varied for each scenario during the five weeks. General trends show the similarities of increasing zooplankton population in the second and the third week, dipping on the fourth week and rebounding on the last week. Specifically, the graphs present that for the 6.0 salinity at 18°C room temperature, the species composition changed as copepods and cladocerans were replaced by the *Keratella* sp., dominating during the final week of the experiment.

Contrary to that, the 6.0 warm scenario showed the sharp increase of copepod nauplii numbers and copepod *Eurytemora* sp. dominated cladocerans and rotifers. Furthermore, the 4.5 warm scenario resulted in an increase in species richness compared to the first week, with noticeable variations in the third week where rotifers dominated. Lastly, for 3.0 scenario, rotifers increased dramatically compared to the initial week.

Differences in Species Richness and Abundance

For zooplankton community, highest mean species richness and abundance come from treatment 6.0W, whereas the lowest mean values were observed in the 3.0 salinity warm treatment.

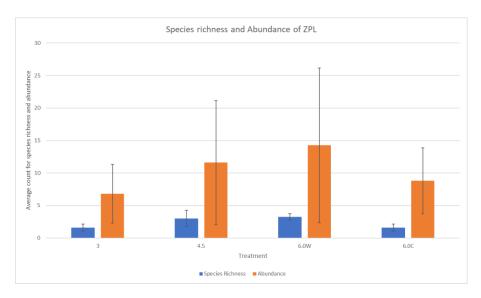


Figure 16. Species richness and abundance of zooplankton in different scenarios. (*Bars represent standard deviation*)

Comparing the community in line with the Climate change effects

The figure above shows how the community composition changed throughout the five weeks and which species thrived in the experiment. With the cold treatment at 6.0psu salinity, a slow and steady shift from copepod-cladoceran community resulted to a complete dominance of the *Keratella* sp.. Increased species richness was observed for both

warmer treatments at 4.5 and 6.0 salinity warm scenarios, suggesting that these conditions were more favourable for the three classes: copepods, cladocerans, rotifers. Moderate changes were observed for low salinity environment at warm temperature. For the 3.0 warm sample, a mix of copepods and rotifers were observed, but with the latter consistently dominating. It is necessary to mention how the species composition changed in the middle of the experiment duration, in week 3. The number of species increased in week 3, followed by a decrease in the following week, and then recovering on week 5. It suggests that the community was able to adapt to the initial stress in week 1 as the count increased with most species surviving.

To see the overall probable effects of climate change on the zooplankton community, we can look at the charts vertically in week 5, starting from 6.0C down to 3.0W. The change in temperature and salinity aims to resemble the impact of climate change in the Baltic Sea as predicted starting from the 6.0 cold scenario, temperature elevates with along with the decrease in salinity. An interesting observation was noted between 6.0°C and 6.0W when the results varied. Having the same salinity treatment at different temperatures, showed a difference in species composition, where the former only had rotifers and the latter had four taxa groups.

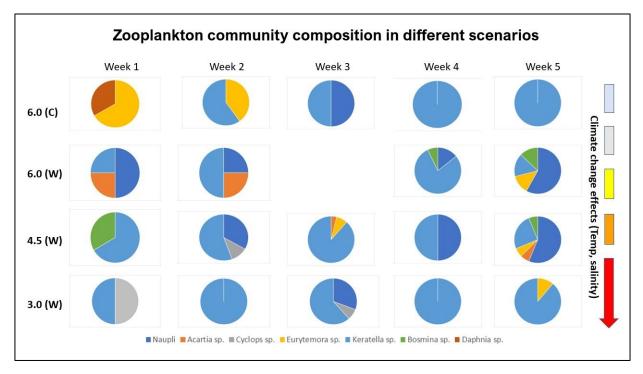


Figure 17. Shifts in community composition throughout the five-week experiment

4.6 Statistical analyses

4.6.1 Analyses for Phytoplankton

ANOVA and T-tests: Trends in Species Richness and Abundance

Statistically, there was no difference in the means of the four different treatments based on the Anova and T-tests conducted.

Table 2. Anova Test for phytoplankton species richness (salinity as a factor)

Anova: Single Facto as a factor)	r for Species					
ANOVA						
Source of						
Variation	SS	df	MS	F	P-value	F crit
Between Groups	0.278571	2	0.139286	0.052561	0.949034	3.982298
Anova: Single						
Factor for						
Abundance						
Source of						
Variation	SS	df	MS	F	P-value	F crit
Between Groups	81.28734	2	40.64367	0.872539	0.442797	3.885294

Table 3. T-test for phytoplankton species richness (Temperature as a factor)

t-Test: Two-Sample Assuming Unequal Variances for Species Richness (Temperature as a factor)

	6.0W	6.0C
Mean	8.25	8
Variance	4.916667	4
Observations	4	5
Hypothesized Mean Difference	0	
Df	6	
t Stat	0.175502	
P(T<=t) one-tail	0.433229	
t Critical one-tail	1.94318	
P(T<=t) two-tail	0.866459	
t Critical two-tail	2.446912	

t-Test: Two-Sample Assuming Unequal Variances for Abundance (Temperature as a factor)

	6.0W	6.0C
Mean	3.53152	5.44502

Variance	17.22405	69.1997
Observations	5	5
Hypothesized Mean		
Difference	0	
Df	6	
t Stat	-0.46025	
P(T<=t) one-tail	0.330771	
t Critical one-tail	1.94318	
P(T<=t) two-tail	0.661542	
t Critical two-tail	2.446912	

Regression Analyses: Observing Relationships between phytoplankton taxa, salinity and temperature

Fluorescence Results

Figure 18 compares the four treatments in terms of fluorescence rates. It was observed that there was an overall decreasing trend for the phytoplankton biomass in all the four treatments. In addition, it can also be observed that in the middle of the experiment, fluorescence recovered following relatively sharp dips the following weeks.

Comparing the fluorescence results to the samples that have been taken for microscopy, the general trends match wherein there was an overall decrease in cell counts from the first week compared to the last week. However, there were observed spikes in the middle of the experiment that were not captured in the microscopy counts.

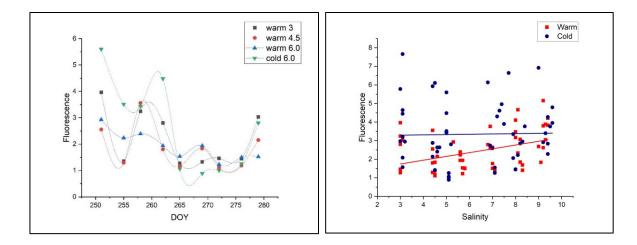


Figure 18. Fluorescence data for the four treatments (Raw data from Lewandowska and Hall, 2022)

In addition, preliminary results from the experiment on fluorescence is shown on Figure 19 using a Linear Regression model. Fluorescence is suggested to have a significant relationship with temperature compared to salinity. The graph represents all the measurements for temperature and fluorescence readings throughout the experiment including in higher salinity range.

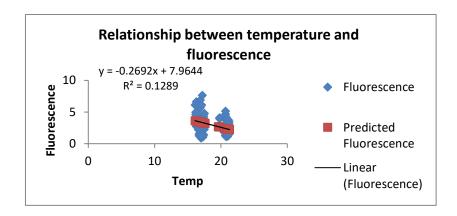


Figure 19. Relationship between temperature and fluorescence (raw data from Lewandoska and Hall, 2022)

Table 4. Regression analysis for Fluorescence

I. Fluorescence and Temperature

	df	SS	MS	F	Significance F	R Square
Regression	1	29.44676	29.44676	15.6796	0.000136	0.12886

II. Fluorescence and salinity

	df	SS	MS	F	Significance F	R Square
Regression	1	5.628426	5.628426	2.676722	0.104791	0.02463

Dinoflagellates: Ceratium spp.

The regression analysis did not show any positive relationship between *Ceratium* spp. and factors salinity and temperature, having the significance value close to 1. It was also observed that throughout the experiment, *Ceratium* sp. was only present in the first week.

Diatoms: Navicula sp., Nitzschia sp., Synedra sp., Fragillaria sp., Thalassiosira sp. and Tabellaria sp.

For diatoms, *Thalassiosira* sp. and *Navicula* sp. showed a significant relationship with salinity as shown in the figure below (Figure 20).

The figure shows the predicted decreasing trend for *Thalassiosira* sp. as salinity increases. Highest cell density for *Thalassiosira* sp. was observed at lower 3 psu salinity, as it slowly decreased towards 6 psu. This is contrary to the predicted trend for *Navicula* sp. where increasing salinity is directly proportional to the total cell density.

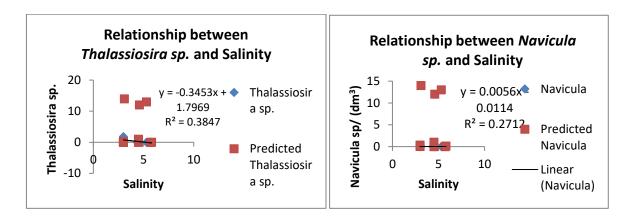


Figure 20. Relationships of *Thalassiosira* sp. and *Navicula* sp. with salinity using LR analysis

Table 5. Regression analyses for *Thalassiosira* sp. and *Navicula* sp.

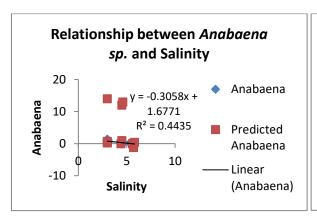
Thalassiosira sp.

πιαιασσιοσπά σφ.							
	df	SS	MS	F	Significance F	R Square	
Regression	1	1.822378	1.822378	7.5012	0.01797	0.384653	
Navicula sp.							
	df	SS	MS	F	Significance F	R Square	
Regression	1	0.000474	0.000474	4.46448	0.056238	0.271158	

Nitzschia sp. on the other hand was observed to have a directly proportional trend with temperature, although statistically it was not found to be strongly significant at 0.08 p-value.

Cyanobacteria: Anabaena sp. Merismopedia sp., and Gomphosphaeria sp.

Results show that there is a significant relationship between *Anabaena* sp. and *Merismopedia* sp. with salinity. The total cell density for *Anabaena sp.* is expected to decrease as salinity increases (Figure 21). For *Merismopedia* sp., an increasing trend is observed for the cell density as salinity increased. On the other hand, *Gomphosphaeria* sp. did not show any significant relationship with salinity and temperature in the regression analyses.



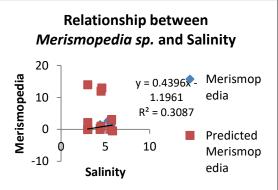


Figure 21. Relationships of *Anabaena* sp. and *Merismopedia* sp. with salinity using linear regression analysis

Table 6. Regression analyses for Anabena sp. and Merismopedia sp.

Regression (Anabaena sp.)

	df	SS	MS	F	Significance F	R Square
Regression	1	1.42914	1.42914	9.56311	0.00932	0.443494

Regression (Merismopedia sp.)

	df	SS	MS	F	Significance F	R Square
Regression	1	2.953312	2.953312	5.357729	0.039143	0.308665

4.6.2 Analyses for Zooplankton

ANOVA and T-tests: Trends in Species Richness and Abundance

Analyses show that the treatments have significant mean differences for species richness for both temperature and salinity scenarios, where alpha = 0.05. In contrast, there were no significant difference observed for abundance in different salinity and temperature scenarios.

Table 7. Anova test for zooplankton species richness (salinity as a factor)

Anova: Single Fac	ies Ric					
Salinity as a factor						
ANOVA						
Source of						
Variation	SS	df	MS	F	P-value	F crit
Between Groups	7.478571	2	3.739286	5.173854	0.026075	3.982298

Table 8. T-test for zooplankton species richness (temperature as a factor)

t-Test: Two-Sample Assuming Unequal Variances: Species Richness with Temperature as a Factor

	6.0(21°C)	6.0(18°C)
Mean	3.25	1.6
Variance	0.25	0.3
Observations	4	5
Hypothesized Mean Difference	0	
Df	7	
t Stat	4.714286	
P(T<=t) one-tail	0.001086	
t Critical one-tail	1.894579	
P(T<=t) two-tail	0.002172	
t Critical two-tail	2.364624	

Regression Analyses: Observing Relationships between Zooplankton taxa, Salinity and Temperature

In relation to the results on species richness, indicating that there were differences found in the different treatments, a linear regression analysis was used to test if salinity and temperature had a relationship on the observed results. Results showed that temperature had a significant p-value (alpha=0.05) to statistically conclude a relationship with the species richness. The trend for temperature and species richness showed a directly proportional result that when temperature increased, species richness also increased, (R^2 = 0.2111, 1 df, 0.047 p value).

Table 8. Regression analysis for species richness and temperature

				Significance		
	Df	SS	MS	F	F	R Square
Regression	1	4.244933	4.244933	4.54996	0.047792	0.211135

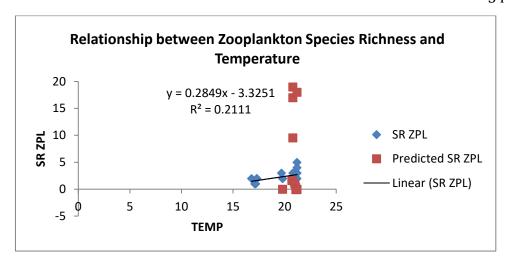


Figure 22. Relationship between species richness and temperature using LR analysis

Linear regression results for salinity and species richness also showed a directly proportional trend, however, the p-value was not significant, and therefore, I cannot fully conclude that salinity had a relationship with species richness, at least for this experiment.

Copepod nauplii

For nauplii, trends show a positive relationship on both temperature and salinity; however, the linear regression analysis did not result in a significant p-value (p-value= 0.175 and 0.083, respectively).

Copepods: Acartia sp, Eurytemora sp. and Cyclops sp.

Different trends were observed for each genera, but no strong significant results were obtained. A positive relationship between the *Acartia spp.* and salinity was observed at alpha 0.089 with an R² of 0.222.

Cladocerans: Bosmina spp.

The regression analysis on *Bosmina spp.* with salinity also showed a positive relationship at 0.058 alpha with a low power of 0.267 R².

Rotifers: Keratella sp.

Keratella sp. did not have a significant relationship with salinity and temperature during the five-week period for the different treatments.

4.6.3 Other observations

Interestingly, *Vorticella sp.* did not show any significant relationship with temperature even though it was present in all warm treatments except for the ambient treatment.



Figure 23. Vorticella sp. observed in warm scenario treatments

5 Discussion

Changes in Phytoplankton Community

For the first research question: "Are phytoplankton groups affected by the temperature and salinity changes related to climate change?", phytoplankton groups did not show any significant mean difference both in species richness and abundance, nor any significant relationships were found with temperature and salinity. It is important to note that that the phytoplankton groups from the four treatments showed similar decreasing general trend (Figures 8,9,10,11) in terms of abundance but they were not statistically significant, potentially due to the small sample size.

However, if we refer to the fluorescence data (Figure 7) that show the measurements for phytoplankton biomass throughout the experiment (including salinity scenarios ranging to 10.5 psu), temperature had a strong relationship with phytoplankton biomass at p-value 0.00. With this, I can infer that temperature was a factor for the direct or indirect changes in the phytoplankton community. Changes in phytoplankton abundance and species richness may be attributed to direct or indirect impacts of temperature including zooplankton grazing (Lewandowska, et.al. 2014). As temperature increases, there are two

possible scenarios that may occur: increased stratification of water, limiting resources of phytoplankton, or stronger grazing of zooplankton which can indirectly affect phytoplankton (Lewandowska, et. al. 2014).

A number of studies about phytoplankton biomass in relation to elevated temperatures were conducted (Suikkanen, et.al. 2013, Lewandowska, et.al. 2014) that have different outcomes. The study from Suikkanen et. al. (2013) suggested that warming resulted to a general increase of phytoplankton biomass towards a community composed of smaller sized groups. On the other hand, the results from Lewandowska's et.al (2014) experiment confirmed field observations where a decrease in phytoplankton biomass was observed as an indirect effect of temperature through increased consumer activity. In addition, one interesting study in boreal lakes explained that phytoplankton biomass decreased during heatwaves due to the decreasing mixed layer depth which phytoplankton depend on for light, and nutrients (Ahonen, et. al. 2023). However, in the context of the Baltic Sea, incidence of warming may increase zooplankton growth, shortening the time lag between phytoplankton and zooplankton peaks, resulting in less phytoplankton in spring (Viitasalo and Bonsdorff, 2022). The cause of decrease in phytoplankton biomass in my study cannot be fully determined, thus further studies on how temperature might have affected these changes directly or indirectly, may be considered in the future.

Looking closely at the genus level, I found interesting observations from *Anabaena* sp. and *Merismopedia* sp., which are included in the cyanobacteria group. Cyanobacteria play an important role in nutrient cycling in the marine ecosystem (Olofsson, et. al. 2020) but some species produce toxins that are considered harmful. Their presence in the Baltic Sea, especially during blooms, have been studied because of their importance, and functional role in the ecosystem, including growth and toxin production. Based on the LR analysis, changes in salinity may affect the growth of these groups as shown in Figure 21. According to the study of Engström-Öst et. al. (2011), growth, including toxin production of *Anabaena* sp. was suggested to be controlled by salinity. Lower salinity environments produced higher growth for *Anabaena* sp., while increased salinity resulted to a significant negative effect on the cyanobacteria (Engström-Öst et. al., 2011). In addition, the adaptation of the cyanobacteria has been studied in line with elevated temperatures (Huisman and Paerl, 2008) and decreasing salinity in the northern Baltic Sea (Olofsson, et.al., 2020).

On the other hand, the total cell density of *Merismopedia* sp., resulted in a directly proportional trend with salinity wherein the increase in salinity further resulted in increased total cell density. A study including the *Merismopedia* sp. in an experiment in New Zealand, conducted by Flöder et. al (2010), determined the positive growth rate of *Merismopedia elegans* with increasing salinity.

Diatoms *Thalassiosira* sp. and *Navicula* sp. also showed significant results in relation to salinity. The former has an inversely proportional relationship with salinity, whereas the latter presented a directly proportional relationship, respectively.

Other genera such as *Nitzschia* sp. showed a positive trend in line with temperature, however, a larger sample size would be necessary to determine the statistical significance of the result.

Overall, Viitasalo and Bonsdorff (2022) concluded that historical trends in the phytoplankton communities have been variable and the global narrative of increased phytoplankton as an effect of climate change can be better assessed by considering factors such as nutrient and carbon dynamics, and food web effects.

Variations in Zooplankton Species Richness

In line with the research question presented for zooplankton communities: "How will the zooplankton communities shift with changes in both temperature and salinity brought about by climate change?", results showed that there was an observed significant difference in terms of species richness in the four treatments where temperature and salinity were analysed as factors.

Based on the linear regression results, species richness is related to the change in temperature, having a significant p-value of 0.04. According to the LR prediction, as temperature increases, species richness will also increase. Temperature influences the structuring of marine ecosystems through its effects on abundance, composition, and trophic efficiency of plankton communities (Richardson, 2008). In the context of species richness, temperature enhances speciation, metabolic rates and restructuring of

community composition may result from species reaching their thermal limits due to elevating temperatures (Bendetti et. al., 2021).

On the other hand, significant mean differences were also observed from different salinity scenarios, showing that salinity could also play a role in zooplankton community shifts. The LR analysis showed a directly proportional trend, however, it has a p-value > 0.05, which is not statistically significant. With this, I cannot fully conclude that salinity had a relationship with the species richness of the zooplankton community in this experiment.

In contrast with our results, a study on zooplankton communities by Helenius et. al (2016) has been conducted in the Gulf of Finland, explaining that salinity is one of the main predictors of both the spatial patterns and functional diversity of zooplankton communities with temperature and turbidity at a lesser extent. However, their study focused on comparing different sites with varying salinity levels in both inner and outer archipelagos in Finland, where other factors may have come into play such as pollution and other anthropogenic activities. While for mesocosm experiments isolating salinity as a factor, it was determined that salinity could be a strong factor in the zooplankton community shift as species have different salinity tolerances (Hall and Lewandowska, 2022).

While we observe variations in species richness based on our results, it is important to note that the warming and freshening of the Baltic Sea do not necessarily result in higher zooplankton diversity. Difference in species richness among different salinity and temperature scenarios could suggest that temperature and salinity are clear potential driving factors for community changes. However, to predict these shifts is not straightforward because other factors at the regional-scale, nutrient dynamics, food web interactions, and anthropogenic effects, which may have direct and indirect effects in the overall composition of the plankton communities (Viitasalo and Bonsdorff, 2022).

Comparing the different scenarios, however, I expect that a warmer and less saline environment at 3.0 psu and 21°C in SW of the Baltic Sea, may result in weaker community shifts, and lower species richness dominated by *Keratella sp*. This supports the results from the mesocosm study of Hall and Lewandowska (2022) suggesting that rotifers will dominate areas shifting to lower salinity due to other species' inability to tolerate less saline

conditions. In addition, rotifers have their optimal conditions at warm and stable water conditions (Viitasalo et al., 1995), which was how the mesocosm was set up.

Zooplankton Abundance and Species dominance

In terms of abundance, no significant difference was determined in the analysis; however, it can be observed from Figure 17, that changes in species composition have occurred and species dominance shifted throughout the five-week experiment. Overall, it was observed that different species dominated in different environmental conditions. Colder temperature at higher salinity (6.0 psu, 18°C) scenario was dominated by rotifers, *Keratella* sp., warmer scenarios at lower salinity levels (6.0 and 4.5) were dominated by copepods (represented by nauplii), whereas lower salinity level (3.0psu) at warmer temperature scenario was dominated by *Keratella* sp. Interestingly, the abundance and dominance of *Keratella* sp. in higher salinity (6.0psu) and lower salinity (3.0) were observed, suggesting its high adaptability to changing environmental conditions. Compared to other species, *Keratella* sp. were found in all scenarios; however, according to Viitasalo et.al (1995), temperature and water stability may have an impact on the abundance of rotifers and cladocerans, rather than salinity. Furthermore, LR analyses did not show any significant relationship between the *Keratella sp.*-temperature, and *Keratella sp.*-salinity, which suggests that there are other factors affecting its presence and growth.

If we analyse shifts more closely from Figure 17, copepod nauplii dominated warmer treatments at higher salinities of 4.5 and 6.0 psu. However, for 6.0 psu cold scenario, a complete shift was observed from copepod-cladoceran to a community entirely dominated by rotifers. It could be that the copepod *Eurytemora* sp. observed in the first two weeks did not have enough food supply because phytoplankton counts for that treatment dropped after the first week, followed by a decreasing trend for the remaining of the experiment. It could also be due to poor adaptability, however, *Eurytemora* sp. is known to be euryhaline brackish genera, along with *Acartia* sp., which can withstand changes in salinity (0.2-12%) (Viitasalo et.al, 1994). In addition, *Eurytemora* sp. was also observed at the lower salinity-warm scenario at 3.0 psu even if the *Eurytemora* sp. from the colder regions have less adaptation potential to warmer temperatures (Karlsson and Winder, 2020). This is an example of how community shifts can also be highly dependent on species level tolerance and preference, making potential variations harder to predict.

6 Limitations of the Study

The mesocosm experiment allowed me to participate from the first day of sampling until the end of five weeks. The major limitation for my research related to the process of identifying and counting the phytoplankton taxa. It was a slow learning curve for the species identification, while counting the cells required many hours. Furthermore, the accuracy may not be perfect, but it was from my best educated knowledge. This forced me to strategize the best way to learn from this research, at the given limited time, and to also maximize my analysis from the samples collected. I decided to implement a modified transect method and focused on the most abundant taxa from the sample. The size of the phytoplankton was also a consideration, as I did not include other smaller groups (except for *Merismopedia sp.*), which are considerably important in community studies. This step may have resulted in variations from my analyses to the actual results generated from general analyses conducted for this experiment, such as the fluorescence data.

In addition, the relatively small sample size can be addressed by collecting a triplicate, to achieve stronger statistical results. A triplicate will also provide some information about the treatment in case a sample is lost.

Finally, this being an indoor mesocosm experiment has the limitations for precisely mimicking the natural conditions such as wave movements, natural light exposure, pH, natural weather conditions, seasonal variations, eutrophication, and other anthropogenic effects. It is important to acknowledge that human error can be present in setting up experiments such as this. However, the methods for setting up the mesocosm were carefully designed and implemented to minimize disturbances and stress to the organisms. The sources of water used for the experiment were selected for its salinity levels, and the process of mixing of these waters, including the gradual addition of salt may have unforeseen effects in the plankton communities, since mixing 0 psu water directly with 5.5 psu may have unnatural effects. The potential induced stress could have probably affected the communities as reflected in the initial low counts for zooplanktons at week one, however, there is no evidence that supports this.

7 Conclusion

For this research, temperature showed a stronger relationship on zooplankton species richness and phytoplankton biomass. However, at the genus level, salinity showed significant relationship with diatoms *Thalassiosira* sp., *Navicula* sp., and cyanobacteria *Anabaena* sp., and *Merismopedia* sp..

Warming sea temperatures being experienced today pose a serious threat to the future landscape of the marine and coastal ecosystems. In terms of taxa observations, it is interesting to note the adaptability of the rotifer *Keratella* sp. in the different scenarios. With the anticipation of freshening and warming of the Baltic Sea, it may be expected to observe *Keratella* sp. thriving and potentially dominating coastal and shallow aquatic ecosystems in southwest of Finland.

Summer blooms may be longer, more intense and more frequent in a warmer sea benefitting phytoplankton, including cyanobacteria (Huisman and Paerl, 2008); however, it is also expected to affect the community composition among groups, and shifts in the dominant species may occur depending on the combined effects of other biotic and abiotic factors.

Looking more closely at the cyanobacteria group, a freshening Baltic Sea suggests the growth for cyanobacteria *Anabaena* sp. (Engström-Öst, et.al, 2011) and a possible decline for *Nodularia* sp.. According to Mazur-Marzec's et.al. (2005) study, *Nodularia* sp. thrives at their optimal growth range of 5-13 psu, and salinity was suggested to be a strong factor to control their development. With the freshening of the Baltic Sea, *Nodularia* sp. could be affected negatively, resulting to a shift in community composition of the cyanobacteria population.

Ciliate protozoan, *Vorticella sp.*, has been observed in warmer temperatures which may suggest that climate change could potentially result to a more conducive environment for them. However, I have not concluded that from this experiment and more research is needed to accept this hypothesis. If this is the case, it is interesting to know the implications of their presence to the plankton communities, and their potential responses to changes in the environment.

Overall, it is expected that climate change will drive changes in the plankton community structure in the Baltic Sea, not only in terms of species dominance but particularly the species richness as well. There is not one determining factor to predict the shifts in the community as both biotic and abiotic factors are at play and affect, not only at the species level, but at the community level. With the complexities set aside, it is overall evident that climate change impacts plankton communities both directly and indirectly, which could transcend at higher trophic levels which depend on these communities.

8 Acknowledgements

I would like to thank my supervisor Jonna Engström- Öst for being a strong motivating force for me to conduct this research and providing me with more than enough support and guidance to finish it. Also, my sincere thanks to Aleksandra Lewandowska and Clio Hall, for allowing me to participate in your mesocosm experiment. It was a huge learning experience that I truly enjoyed. I also want to thank all the staff of Tvärminne Zoological Station for allowing me to use your facilities, sediment chambers, and microscope for this research. Lastly, special thanks to my friends, Jinzhu Su and Iris Orizar, for providing support and guidance throughout this study.

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