APPENDIX 3

TA PROJECT REPORT (TEMPLATE)

(see following pages)

TA PROJECT REPORT PACKAGE

- The completed and signed forms included in this package should be sent by email to jerico.ta@marine.ie and **jerico**-s3**@ifremer.fr** within **one month after the completion of the TA project** by the User Group Leader.
- **Refunding of the TA reimbursement to the user group will be processed as soon as these forms will be submitted**.
- The TA project report will be published in the JERICO-S3 website. The report, as well as other information collected with the attached forms, will be used to report to the European Commission.
- **Please note that any publication resulting from work carried out under the JERICO-S3 TA activity must acknowledge the support of the European Commission – H2020 Framework Programme, JERICO-S3 under grant agreement No.871153.**

1. Project Information

Proposal reference number[1]	22/1002919
Project Acronym (ID)[2]	BalHObEx
Title of the project[3]	**Baltic Sea Heat Waves: Observation and Experimentation**
Host Research Infrastructure[4]	**SYKE MRC-lab and Alg@line**
Starting date - End date[5]	SYKE MRC-lab 19/8/2022 to 4/9/2022 Algaline 29/6/2022 to 21/9/2022 (8 sampling dates)
Name of Principal Investigator[6] **Home Laboratory** **Address** **E-mail address** **Telephone**	Iordanis Magiopoulos Institute of Oceanography, Hellenic Centre for Marine Research Ex-American Base iordanis@hcmr.gr +30 6988 55 22 25

2. Project objectives[7] (250 words max.)

BalHObEx project followed a holistic approach to study the effects of extreme heat waves on the planktonic food web of the Baltic Sea.

More specifically:

1. **Investigate the effects of extreme heat waves on the marine plankton food web via a mesocosm experiment**. Mesocosms allow experimentation on whole plankton food webs in close to real-life conditions and they are considered the most reliable way to test hypotheses and predict effects of environmental pressures on the complex marine ecosystems. Therefore, the participation of the BalHObEx research team to the already planned mesocosm experiment at the SYKE mesocosm facility allowed the study of the effects of the elevated temperature (from 16℃ up to 22℃) on the

[1] Reference number assigned to the proposal by the TA-Office.

[2] User-project identifier used in the proposal.

[3] Title of the approved proposal. The length cannot exceed 255 characters

[4] Name of the installation/infrastructure accessed with this project. If more than one installations/infrastructures are used by the same project, please list them in the box.

[5] Specify starting and end date of the project (including eventual preparatory phase before the access).

[6] Fill in with the full contact of the Principal Investigator (user group leader).

[7] Write the short-term, medium and long-term objectives of the project. Use no more than 250 words.

microbial community under controlled environmental conditions and in combination with a number of various other analytical tools and real-time data from sensors, collected by other participating researchers.

2. **Compare and combine the results from the above mentioned mesocosm experiment with findings in the natural environment** in order to get a more complete view on the effects of the heat waves on natural plankton communities of the Baltic Sea.

3. Main achievements and difficulties encountered (250 words max.)[8]

Both the participation in the mesocosm experiment and the in-situ samplings using the FerryBox facility of SYKE went according to the plan.
The BalHObEx team and the scientists and technicians from the SYKE facility had an excellent collaboration from the planning of both the experiment and Ferrybox samplings to the shipping of the samples back to HCMR.
The samples collected during the mesocosm experiment were analysed at HCMR in Crete, Greece and the data collected will be combined with data generated by other teams, such as the pigment content (by the CNRS-MARBEC team) and bacterial production (by the SYKE team), to produce concrete conclusions on the responses of marine microbes to sudden temperature increases.

The only difficulty encountered was the shipment of samples from Helsinki, FI to Heraklion, GR, due to their nature (chemically fixed samples in dry ice) and the consequently high shipping cost.

4. Dissemination of the results[9]

The activity was presented on the JERICO-RI web page (https://www.jericori.eu/2022/09/06/joint-jerico-s3-and-aquacosm-plus-study-on-baltic-sea-heatwaves/) and on the AQUACOSM-plus web page (https://www.aquacosm.eu/news/article/joint-aquacosm-plus-and-jerico-s3-study-on-baltic-sea-heatwaves). In addition, a news article with a more “personal” and “family” perspective was published in the AQUACOSM website, showing the challenges faced by a scientist that wants to take part in a TA activity but is also a parent of a very young kid (https://www.aquacosm.eu/news/article/participation-in-a-mesocosm-experiment-at-syke-helsinki-hcmr-team-and-little-maria). All articles were promoted through AQUACOSM-plus social media.

[8] Describe briefly the main achievements obtained and possible impacts, as well as possible difficulties encountered during the execution of the project. Use no more than 250 words.

[9] Describe any plan you have to disseminate and publish the results resulting from work carried out under the Transnational Access activity in JERICO -S3: scientific articles, books - or part of them -, patents, as well as reports and communication to scientific conferences, meetings and workshops. Highlight peer-reviewed publications. **Note that any publications resulting from work carried out under the JERICO -S3 TA activity must acknowledge the support of the European Commission – H2020 Framework Programme, JERICO -S3 under grant agreement No. 871153.**

The results from both the experiment and the field sampling will be presented in scientific meetings and will be published in scientific peer-reviewed journals in collaboration with SYKE and the other teams on location.

5. Technical and Scientific preliminary Outcomes (2 pages max.)[10]

During the mesocosm experiment, samples for the analysis of the microbial community were collected from the beginning (Day 0 – just before the temperature increase) until the end (Day 9) of the experiment. The temperature treatments were 16℃, 18℃, 20℃ and 22℃.
For the field samplings, samples were collected from 4 stations (St 19, 21, 22 and 24 where St 24 was just outside the port of Helsinki, St19 was a relatively open water station - N.B. the water for the mesocosm experiment was collected in the area between St 21 ad St 22) at 8 time points. The first sampling was conducted on the 29th of June and the last on the 21st of September. During that period, *in-situ* surface water temperature ranged from approx. 15-16.5℃ (June 26th) to 21.5℃ (August 19th) and went back to 15℃ (September 21st).
The BalHObEx team collected samples for the analyses of viruses (abundance and cytometric groups) and viral production, heterotrophic and autotrophic bacteria (abundance, cytometric groups, pigments per cell, relative size), autotrophic nano-flagellates (abundance, pigments per cell, relative size) and microplankton (abundance and diversity) from both the mesocosm experiment and the FerryBox samplings (apart from viral production in the field samplings as it was not technically possible).
All samples were shipped to HCMR, Crete Greece and were analyzed by means of flow cytometry (using a BD FACSCalibur and a Life Technologies Attune NxT flow cytometer) and inverted microscopy (using an Olympus IX70 inverted microscope).

Bellow some of the above mentioned data will be presented briefly. More data analysis will follow, in close collaboration with the teams that participated in the mesocosm experiment as well as with the SYKE team that operates the FerryBox.

Heterotrophic Bacterial abundance (Figure 1) didn't change significantly between treatments at the beginning of the experiment, with only a significant increase at the +2℃ treatment (18℃) on Day 3. By the end of the experiment, bacterial abundance was higher at the 20℃ treatment.

[10] Describe in detail results and main findings of your experiment at the present stage.

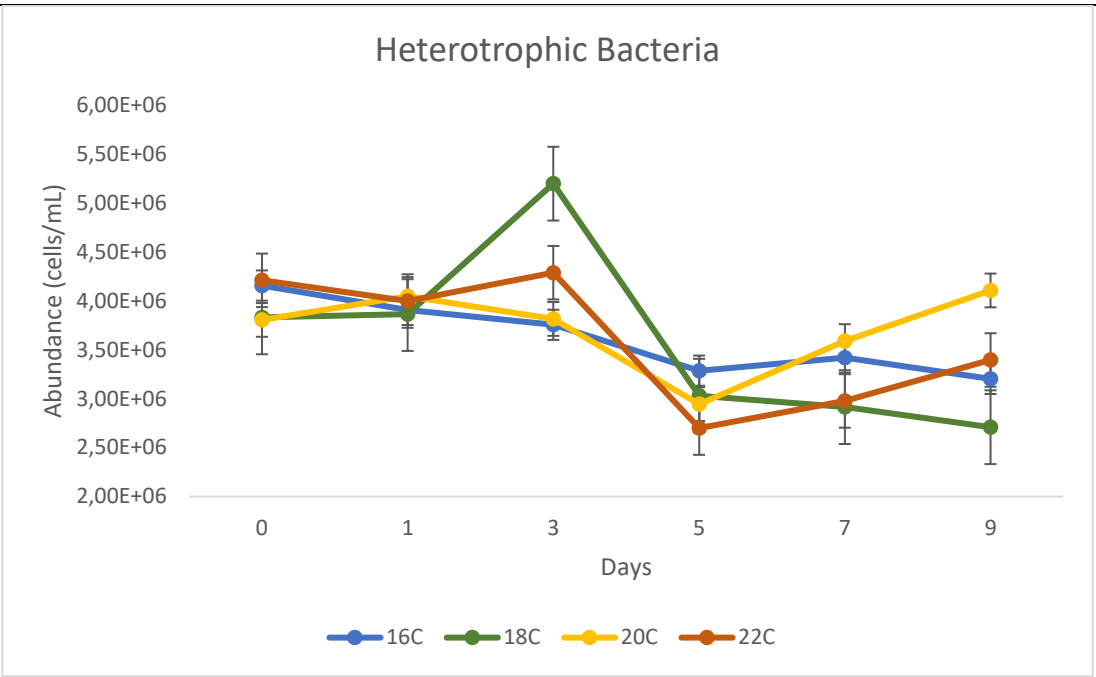


Figure 1. Abundance of heterotrophic bacteria (in cells/mL) during the mesocosm experiment.

Also, the abundance of autotrophic bacteria (Figure 2), which were mainly belonging to the *Synechococcus* genus, was higher in the 20 and 22℃ treatments at the end of the experiment.

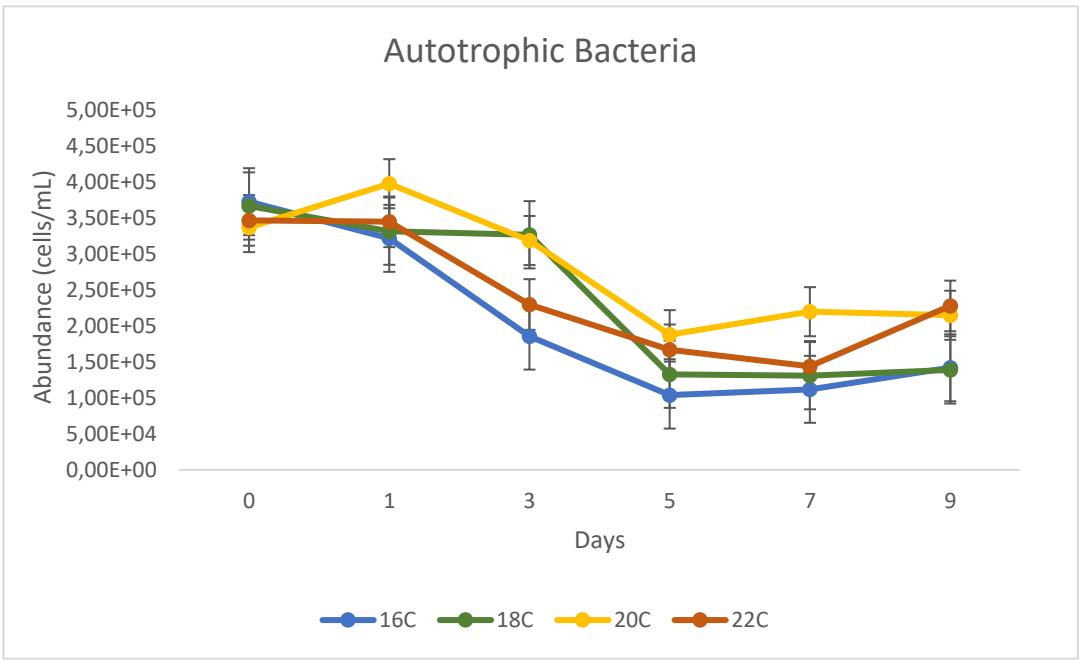


Figure 2. Abundance of autotrophic bacteria (in cells/mL) during the mesocosm experiment.

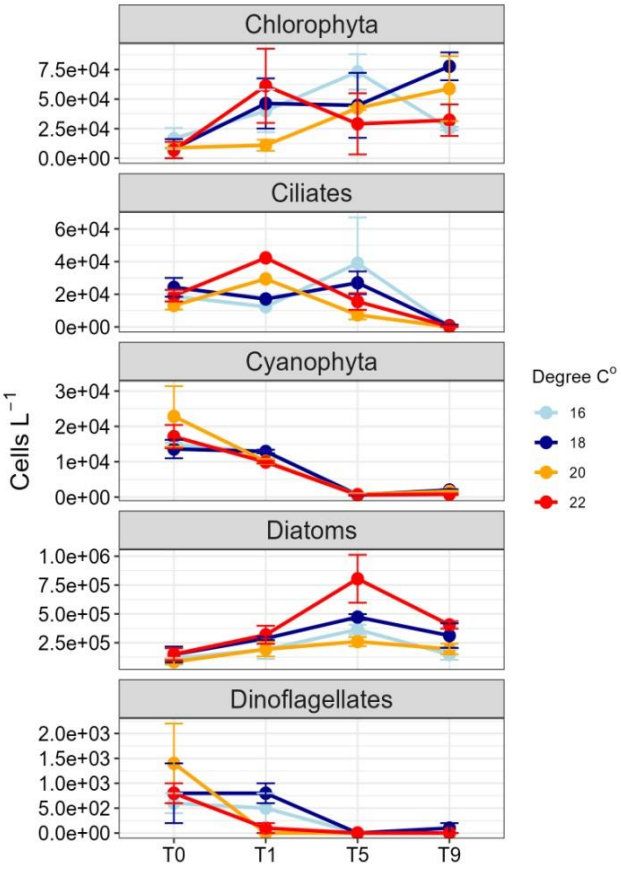


Figure 3. Abundance of the main microplankton groups (in cells/L) during the mesocosm experiment.

From the analysis of the microplankton community (Figure 3), it was found that Chlorophyta responded negatively at 20℃ in the beginning of the experiment while Ciliate abundance increased at the two higher temperature treatments in the same time point. From the middle of the experiment until the end, no statistically significant changes were found, regarding the microplankton abundance.

The results of the field samplings showed that Heterotrophic Bacterial abundance, from all stations, (Figure 4) increased along with the temperature increase (from June 26th to August 19th when the maximum temperature was recorded); however, it was not significantly different after August 31st and onwards (when temperature decreased to 18 and 15℃), while during the same period, the contribution of the High Nucleic Acid (HNA) bacteria to the total bacterial community decreased from approximately 35-45%, between the end of June to mid-August, to 25-30%, between the end-August and the end-of September.

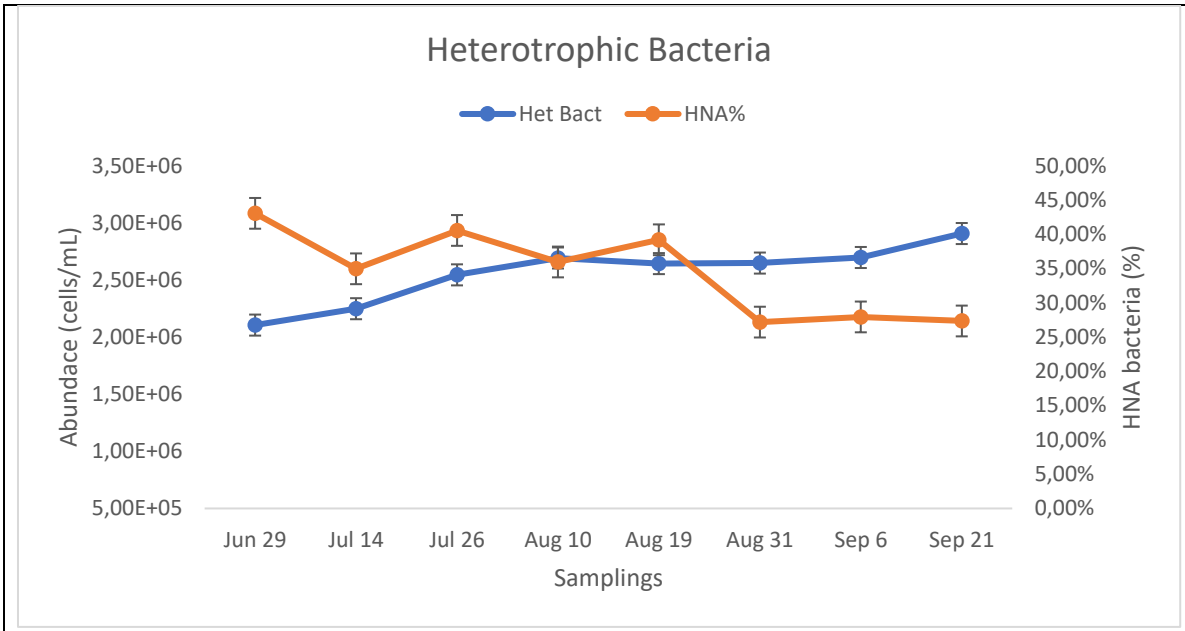
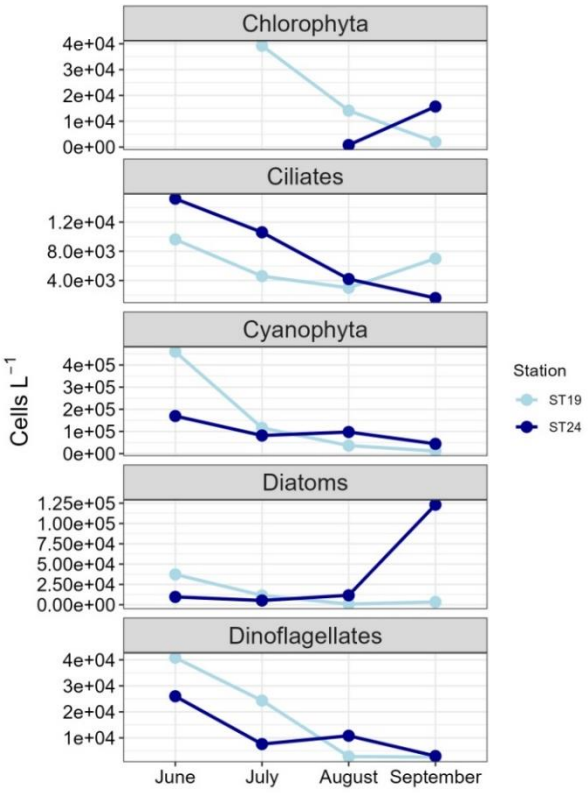


Figure 4. Heterotrophic Bacterial (Het Bact) Abundance (in cells/mL) and High Nucleic Acid (HNA) bacterial relative abundance (in %) to the total bacterial community from the field samplings. Data presented here are the average of all 4 sampling stations.



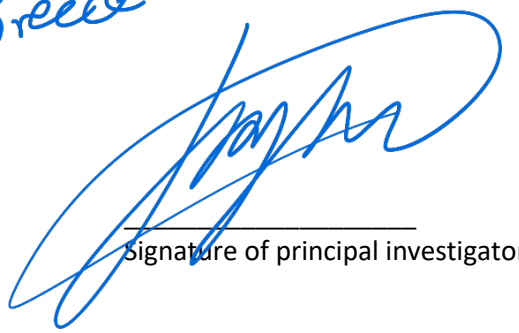
The analysis of the microplankton community showed a decrease in abundance of most microplankton groups (mainly Ciliates and Dinoflagellates) from June to September, with the exception of Diatoms and Chlorophyta at the stations next to the Helsinki Harbor during the last sampling mission.

Figure 5. Abundance (in cells/L) from the main microplankton groups from Stations 19 (open water) and 24 (Helsinki port) during the field samplings

The Raw data that were obtained after the mesocosm experiment, by analysing the samples using the flow cytometers and microscopes at HCMR-Crete, **will be available on open source** after quality checking and cross checking with another lab at HCMR-Athens and will be part of an open access paper describing the effect of a simulated heat-wave on the plankton community and size distribution. The Metadata and Data will be available on open source in an open source publisher (for example Pangaea https://pangaea.de and/or the AQUACOSM Data/Metadata Portal https://www.aquacosm.eu/metadata-and-data) and on the scientific journal site (if it is available such option).

Heraklion, Crete, Greece
4/9/2023

Location and date



Signature of principal investigator