METEOR-Berichte

Eastern Mediterranean Sea – Process Study

Cruise No. M197

30.12.2023–06.02.2024 Limassol (Republic of Cyprus) – Catania (Italy) EMS-PS

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1 Cruise Summary

1.1 Summary in English

The research cruise M197 with the RV METEOR sailed January $6th 2024$ to February $6th 2024$ from Limassol (Republic of Cyprus) to Catania (Italy), with a focus on investigating the seawater and sediment biogeochemistry in the Eastern Mediterranean Sea. The two specific foci of the research cruise were to (i) investigate the physical, chemical and biological factors regulating the productivity and sinking carbon flux in this region, and (ii) investigate natural and human induced changes in the region over the last few thousand years. On the research cruise, 30 stations were occupied. Seawaters were collected from the surface to the seafloor to measure chemical properties of the seawater, including major and trace nutrients, the carbonate system, microplastics, and the microbial communities inhabiting the water column. Experiments were conducted to assess rates of primary production and nitrogen fixation, and the nutrients regulating phytoplankton growth. Sinking carbon fluxes were determined using the thorium-234 approach. The types and abundance of zooplankton were determined via various nets and two camera systems. The deposition flux and chemistry of aerosols was also assessed. Sediment cores were collected to determine nutrient fluxes into the overlying water column as well as investigate past environmental conditions. Collectively our research will provide an important advance in the network of factors that regulate the chemistry and biology of this system and inform ocean biogeochemical models of this region to make more realistic predictions of climate change induced impacts.

1.2 Zusammenfassung

 Die Forschungsfahrt M197 an Bord von RV METEOR führte vom 6. Januar 2024 bis zum 6. Februar 2024 von Limassol (Republik Zypern) nach Catania (Italien) und konzentrierte sich auf die Untersuchung der Biogeochemie des Meerwassers und der Sedimente im östlichen Mittelmeer. Die beiden Schwerpunkte der Forschungsfahrt waren (i) die Untersuchung der physikalischen, chemischen und biologischen Faktoren, die die Produktivität und den sinkenden Kohlenstofffluss in dieser Region regulieren, und (ii) die Untersuchung der natürlichen und vom Menschen verursachten Veränderungen in der Region während der letzten paar tausend Jahre. Auf der Forschungsfahrt wurden 30 Stationen besetzt. Das Meerwasser wurde von der Oberfläche bis zum Meeresboden gesammelt, um die chemischen Eigenschaften des Meerwassers zu messen, einschließlich der Haupt- und Spurennährstoffe, des Karbonatsystems, des Mikroplastiks und der mikrobiellen Gemeinschaften in der Wassersäule. Es wurden Experimente durchgeführt, um die Raten der Primärproduktion und der Stickstofffixierung sowie die Nährstoffe, die das Wachstum des Phytoplanktons regulieren, zu bewerten. Die sinkenden Kohlenstoffflüsse wurden mit Hilfe des Thorium-234-Verfahrens bestimmt. Die Arten und die Abundanz des Zooplanktons wurden mit verschiedenen Netzen und zwei Kamerasystemen bestimmt. Der Depositionsfluss und die Chemie der Aerosole wurden ebenfalls untersucht. Es wurden Sedimentkerne entnommen, um die Nährstoffflüsse in die darüber liegende Wassersäule zu bestimmen und die Umweltbedingungen der Vergangenheit zu untersuchen. Insgesamt werden unsere Forschungsarbeiten einen wichtigen Beitrag zum Netzwerk der Faktoren leisten, die die Chemie und Biologie dieses Systems regulieren, und die biogeochemischen Ozeanmodelle dieser Region informieren, um realistischere Vorhersagen über die Auswirkungen des Klimawandels zu machen.

2 Participants

2.1 Principal Investigators

2.2 Scientific Party

3 Research Program

3.1 Description of the Work Area

Hydrography- The Mediterranean Sea is a semi-enclosed marginal basin that is separated into two parts by the Straits of Sicily (with a saddle depth of 430 m): Eastern and Western Mediterranean Seas. The Western Mediterranean is in direct contact with the North Atlantic at the Strait of Gibraltar which has a sill depth of 284 m. Here, North Atlantic waters flow into the Mediterranean as surface waters, and Mediterranean deep waters form a return flow. The Mediterranean is a 'negative' basin with respect to water balance with evaporation exceeding precipitation and run-off. There is also a net cooling within the Mediterranean and the outflow is hence denser (cooler and saltier) than the inflow. As a consequence, the Mediterranean is well ventilated to the bottom. The Eastern Mediterranean has complex circulation patterns that evolve at several different spatio-temporal scales, with the various sub-basins (Levantine Sea, Aegean Sea, Ionian Sea, and Adriatic Sea) dynamically interacting and determining the general hydrology of the overall basin. The circulations in the Adriatic and Aegean Seas are cyclonic, but in the Levantine Basin the situation is more complex with cyclonic sub-basin scale gyres north of the mid-Mediterranean jet and anticyclonic gyres of Mersa-Matruh north of Libya and Shikromona close to Israel to the south. The mean horizontal circulation in the Eastern Mediterranean at intermediate and deep levels is cyclonic, similar to the surface circulation. In the vicinity of the cyclonic Rhodes Gyre, Levantine Intermediate Water (LIW) is formed which spreads cyclonically at mid-depth (200-600 m) and is the source of deep water in the eastern as well as Western Mediterranean which ultimately forms the deep-water outflow at Gibraltar. The deep-water formation areas in the Western Mediterranean can be found in the Gulf of Lions, but also in the Eastern Mediterranean in the Adriatic and Aegean Seas (Eastern Mediterranean Deep Water (EMDW)). The other water masses in the Eastern Mediterranean include warm and saline Levantine Surface Water (LSW) which is formed in the Levantine Basin in summer, and the low salinity Atlantic Water (AW) in the subsurface which enters the Eastern Mediterranean from the west. Salinities over 39.1 can be found in the surface waters of the Eastern Mediterranean. Even at 4000 m depth, bottom water temperatures are above 12.5°C with clear evidence of warming (see below). Deep water oxygen levels exceed 200 μmol/kg.

3.2 Aims of the Cruise

 The overarching aim the EMS-PS programme is to use the rapidly changing Eastern Mediterranean Sea (EMS) as a natural laboratory to gain mechanistic understanding of biogeochemical and ecosystem transitions of a future (sub-)tropical ocean affected by global warming and other anthropogenic pressures. The M197 cruise on RV METEOR had the following objectives: (i) characterizing nutrient biogeochemistry and phytoplankton nutrient limitation of seawaters in the Eastern Mediterranean Sea, (ii) documenting in detail the microbial communities that inhabit these waters from the surface ocean to sediments, (iii) assessing mechanistic connections between nutrient biogeochemistry, surface ocean productivity, deeper water metabolism, and shelf sediments in the cycling of carbon and major nutrients, (iv) using the sedimentary record to assess past environmental change in the EMS.

Our observations on the M197 research cruise will be used alongside existing autonomous platforms in the EMS and satellite observations to document spatial/temporal change.

3.3 Agenda of the Cruise

 On the research cruise, 30 stations were occupied where a programme of deployments were undertaken. Seawaters were collected from the surface to the seafloor to measure chemical properties of the seawater, including major and trace nutrients, major elements, the carbonate system, microplastics, and the microbial communities inhabiting the water column. Experiments were conducted to assess rates of primary production and nitrogen fixation, and the nutrients regulating phytoplankton growth. Sinking carbon fluxes were determined using the thorium-234 approach. The types and abundance of zooplankton were determined via various nets and two camera systems. The deposition flux and chemistry of aerosols was also assessed. Sediment cores were collected to determine nutrient fluxes into the overlying water column as well as investigate past environmental conditions. Collectively our research will provide an important advance in the network of factors that regulate the chemistry and biology of this system and inform ocean biogeochemical models of this region to make more realistic predictions of climate change induced impacts. All deployments generally functioned successfully, except for the multicore sediment coring device where the closing mechanism initiated early on a number of deployments undertaken in stronger swell (ca. 2-2.5 m) that led to transient increased tension on the deployment wire. All research activities were carried out in the frame of the OSPAR Code of Conduct for Responsible Marine Research in the Deep Seas and High Seas of the OSPAR Maritime Area. No explosive or noise intensive measurements were conducted and all chemicals used onboard were returned to home laboratories.

Fig. 3.1 Track chart of RV METEOR Cruise M197. Solid dots are station locations.

4 Narrative of the Cruise

The departure of research cruise M197 was delayed one week, with departure from the port of Limassol, Republic of Cyprus, on 06.01.2024 rather than intended date of 30.12.2023. This was due to unforeseeable delays in delivery of equipment containers containing essential equipment for the research activities.

We departed port at ca. 09:30 local time (07:30 UTC) on 06.01.2024 and deployed the towed water sampling device on entering the first working area. Hydroacoustic observations were collected through the night whilst in working areas. Station 1 started at 06:00 ship time (04:00 UTC) on 07.01.2024. In general, all operations were successful; however, the in-situ pumps pumped only a very small volume of seawater and the multinet zooplankton net did not close on first attempt; the latter was repeated and functioned successfully. Between 07.01.2024 until 11.01.2024 a series of four stations were conducted across the so-called 'Cyprus eddy', an anticyclonic circulating, semi-permanent oceanographic feature to the south of the Republic of Cyprus. On 08.01.2024 the reason for the misfunctioning in situ pumps was identified and from thereon they functioned well. On 09.01.2024 the towed video camera system ('PELAGIOS') lost power ca. 1 hour into the 3.5-hour deployment; the LED light usage was reduced and this solved the problem. In the night of 09.01.2024 there was some rain and lightning with a swell of 1-2 m. On 10.01.2024 the closing mechanism on the multicore sediment coring device triggered early and, as a result, no sediment core was achieved. This was identified to be due to the slightly elevated swell (ca. 2 m) that increased tension on the wire periodically and triggered the closing mechanism.

On 10.01.2024 two scientists tested positive for Covid-19 after feeling a little sick. At that point, procedures were taken to reduce the chance of the infection spreading; specifically, the infected scientists wore masks in public areas and ate meals at different times and all scientists were provided with antigen Covid-19 antigen tests to take over the next 5-day period. On 11.01.2024 and 12.01.2024 one additional scientist on each day tested positive for Covid-19. On 11.01.2024 and 12.01.2024 the swell remained elevated (ca. 2 m) and the multicore device closed prematurely. On 13.02.2024 a station was occupied over Eratosthenes Seamount. The swell had calmed to ca. 1.5-2 m and the multicore deployment was successful. On 14.01.2024, Station 8 was conducted to the north of Eratosthenes Seamount. The swell remained low (1- 1.5 m) and the multicore deployment was successful. From this date onwards no more scientists were positive with Covid-19.

On 15.01.2024 we had our last station in the Republic of Cyprus EEZ and sailed west to the Greek EEZ. The passage between them was slowed by strong winds and swell (ca. 3 m) against the ship and an engine problem that needed repair; as a result, we arrived on station ca. 4 hours later than planned. Between 16.01.2024 to 04.02.2024 we progressed steadily to the northwest occupying stations each day starting between $06:00 - 08:00$ local time $(04:00 - 06:00$ UTC) and finishing between ca. $22:00 - 03:00$ local time $(20:00 - 01:00$ UTC).

On 19.01.2024 at ca. 17:30 ship time (15:00 UTC) the 75k Hz ADCP was replaced with the Posidonia USBL receiver for accurate underwater positioning of the multicore at the next station, which was overlying the Napoli Mud Volcano, the crater of which contains a brine

pool. Three stations were conducted around this site with a total of two multicore deployments, two deployments of the trace-metal-clean CTD, and both day and night time deployments of zooplankton nets (with night-time-only deployments at the majority of the other stations). In the evening of 20.01.2024 we had a barbecue on the working deck to mark the approximate mid-point of the research cruise. During the evening of 20.01.2024 there was a notification sent from a nearby vessel that a small boat (likely containing refugees) had been sighted, which, given the expected upcoming bad weather, might have needed assistance. The RV METEOR sailed in the direction of the small boat in the chance that assistance from might need to be delivered. Throughout the evening updates were followed and, in the end, another vessel provided assistance and the RV METEOR returned to the previous station site.

On 22.01.2024 the intended route was adjusted to move north towards Crete, in order to shelter from strong wind and waves from the north. The swell still remined 1.5 m going on 2.5- 3 m at Station 17 and the multicore was deployed but closed early and was recovered to deck and a second deployment was not attempted. On the night of 22.01.2024-23.01.2024 we had extra time (due to the failed multicore) and used this to cross the cyclonic eddy to the south to acquire additional hydroacoustic data before occupying Station 18 at the site of a deep canyon around 60 km south of Crete. At 11:00 ship time (10:00 UTC) we carried out a live video call with a group of school children, describing our scientific programme and life on board a research vessel. The school children were taken on a tour through the ship from the bridge, the weather station, the ships gym and hospital, through to the working deck and the scientific laboratories. Apparently, the school group found it interesting and entertaining and hopefully inspired them a little in the subject of oceanography. Wind and waves calmed significantly throughout the day.

On 28.01.2024, the swell $(1.5 - 3 \text{ m})$ and wind $(4 - 9 \text{ Bft})$ increased rapidly during the day and a waterspout sighting was made at ca. 08:00 ship time (07:00 UTC). At ca. 15:00 ship time (14:00 UTC) it was decided that it was too rough for deployment of the in-situ pumps and multicore. Due to rough swell, the multicore shut prematurely at Stations 24 and 25 (29.01.2024 and 30.01.2024, respectively), whilst other deployments proceeded without problems.

On 31.01.2024 we occupied our deepest site with ca. 5100 m water depth and the station was conducted with calm sea conditions and sunny weather. A second live video call with school children was conducted; this time 30 classrooms joined with ca. 1000 school children viewing in total. Between 01.02.2024 and 04.02.2024 we conducted our final four stations of the cruise in calm sea conditions (ca. 0.5-1.5 m swell) and pleasant weather. The final station on 04.02.2024 (Station 30) only comprised of standard and trace-metal-clean CTDs to a depth of ca. 500 m in the morning and zooplankton nets in the evening. The remainder of the day was used for final sample processing and packing. On 05.02.2024 the scientific team with the help of the ship's crew packed equipment and cleaned laboratories. On 06.02.2024 we arrived in Catania (Italy) at ca. 09:00 local time where shipping containers were unloaded from the vessel and frozen samples were handed over to a courier company. Two scientists left the vessel on the afternoon of 06.02.2024 and the remainder on the morning of 07.02.2024.

5 Preliminary Results

5.1 Stainless-Steel CTD Surveying

(H. Melzer, P. Damke)

To study the water column, the RV METEOR's stainless-steel rosette (SS-CTD) was deployed during clean-ship conditions. The SS-CTD was equipped with a *Sea-Bird Scientific SBE 911Plus* instrument, 24 Niskin bottles, an Underwater Vision Profiler 5 DEEP device (UVP) as well as a nitrate sensor from *TriOS GmbH*. The SBE 911Plus device measured seawater temperature (2 sensors - Ser. Nr. 5283 and 5655) conductivity (2 sensors – Ser. Nr. 4058 and 3717) and pressure (Ser. Nr. 1497). Additionally, a sensor for dissolved oxygen was attached to the CTD. However, the oxygen sensor showed a strong drift and offset, therefore it was replaced after station 15 (file number 'met_197_1_042') from Ser. Nr. (2415) to (2337). The latter sensor was taken from the TM CTD. Therefore, the data from the TM CTD from the same station can be used for analyses. Further, fluorescence and turbidity were measured by an instrument from *Wet Labs* (Ser. Nr. 2718) and PAR/ SPAR was recorded by an instrument from *Biospherical Instruments Inc* (Ser. Nr. 20353)*.* The PAR sensor from the ship CTD could not be deployed for depths greater than 2000 m, therefore it was replaced by a sensor from GEOMAR (Ser. Nr. 70714). Note, that the PAR data is not available for the first cast.

The deck unit was operated using the setup file 'M197_hm.psa' with the config file 'M197_mit-PAR-GEOMAR.xmlcon'. The file name convention was 'met_197_1_nnn'. The data were processed to remove spikes and then a moving mean smoothing was applied, binning the data to 10 m segments. The data was processed and plotted using the program *MATLAB*, and the raw data was converted into an ASCII format using *Sea-Bird Scientific's SBE Data Processing* program. In total 89 CTD profiles were recorded. The first deployment took place on January 07th 2024 and the last CTD took place on February 04th 2024.

5.2 Trace Metal CTD Surveying

(D. Jasinski, A. Nicolas, H. Melzer, P. Damke, Y. Ruan)

In addition to the RV METEOR's stainless-steel rosette, a second CTD for trace metal sampling (TM CTD) was deployed also during clean-ship conditions. The TM CTD is made with a titanium frame and was operated via a separate containerized winch equipped with a Kevlar cable. The TM-CTD was equipped with a *Sea-Bird Scientific SBE 911Plus* instrument and 24 Niskin bottles. The SBE 911Plus device measured seawater temperature (2 sensors - Ser. Nr. 5562 and 5661) conductivity (2 sensors – Ser. Nr. 4064 and 4084) and pressure (Ser. Nr. 1086). Additionally, two sensors for dissolved oxygen were attached to the CTD. However, one of the sensors was broken and got replaced after the first cast Ser. Nr. (1718) to Ser. Nr. (2336). The secondary sensor (Ser. Nr. 2237) was working fine, nevertheless, due to a malfunction of the oxygen sensor on the SS CTD it got taken off at station 15 and attached to the SS CTD (after filenumber 'met_197_1_tm_015'). Furthermore, it included a transmissometer from *WET Labs* (Ser. Nr. CST-1520DR). The deck unit was operated using the setup file 'Seasave_TM_METEOR_197_hm.psa' with the config file 'MET 197 1 tm hm.xmlcon.'

The data were processed to remove spikes and then a moving mean smoothing was applied, binning the data to 10 m segments. The data was processed and plotted using the program *MATLAB*, and the raw data was converted into an ASCII format using *Sea-Bird Scientific's SBE Data Processing* program. In total 29 CTD profiles were recorded. The first deployment took place on January 07th 2024 and the last CTD took place on February 04th 2024.

5.3 Underway Sampling from a Towed Water Sampling Device

(A. Nicolas, B. Ankri, Y. Han, Y. Ruan, A. Blachinsky, J. Ivaldi)

In order to have a better overview of the surface biogeochemical processes, we used a towed, trace-metal clean sampling device (so-called tow fish) used for pumping (teflon double diaphragm pump, Dellmeco) surface water (ca. 3-5 m depth) via acid-washed tubing. Samples were collected in a TM clean bench (plastic sheeting enclosing a laminar flow hood). Sampling took place in between stations, when possible, with a variable time resolution.

All samples were collected using a filter capsule (AcroPak 500 Supor Membrane 0.8/0.2 μm). **Dissolved TM:** Filtered water samples were collected into acid clean 125 mL LDPE bottles (rinsed 3 times with sample before collection). Samples were acidified with 175 μL *Optima* grade HCl (see section: Trace metals sampling and analysis). **Major elements**: Filtered water samples were collected into acid clean 5 mL plastic vials (rinsed 3 times with sample before collection). (See: Major elements sampling and analysis section). **Inorganic/organic macronutrients:** Samples were collected into duplicate acid clean plastic 15 mL (inorganic) or single 50 mL (organic) vials (rinsed 3 times with sample before collection). For inorganic nutrients, one sample was collected for nano- and one for micromolar concentrations of inorganic macronutrients, the latter measured on board by A. Mutzberg (See: Macronutrients section). Nanomolar nutrient samples were immediately frozen at –20 °C and will be analyzed in GEOMAR.

5.4 Salinity

(M. Liadova, P. Damke)

Salinity values of the different CTD casts were measured by salinity sensors attached onto the rosette frames. There were two different CTD types: The stainless steel CTD (SS CTD) and the trace metal clean CTD (TM CTD). 159 discrete salinity samples were collected into 0.33 mL glass bottles from different depths of the water column and the CTD casts to calibrate these sensors. For Station 1, 4 samples were taken in TM CTD cast. For Stations 2 and 3, 6 samples were taken in TM CTD cast. For the SS CTD, 3 samples were taken in the deep cast and 1 in the shallow cast. From Stations 4, 5 or 6 samples per station were taken following the order: (1) 3 samples from TM CTD, 1 or 2 samples from the deep cast of SS CTD, and 1 sample from the shallow cast of SS CTD, or (2) 2 samples from TM CTD, 2 sample from deep SS CTD, and 1 sample from shallow SS CTD. In total, three depths were covered per station: the deepest point, a shallow depth at around 80-40 m depth, and a mid-water column depth. Salinity values will be analyzed after the cruise at GEOMAR. See also Sections 12.01–12.02.

In addition to the salinity samples from the CTD casts, a number of samples from the ship's dual thermosalinograph (TSG) were taken at the outlet MMC2. Most of the samples were taken underway, some of them on station. The samples will be analysed after the cruise at GEOMAR. See Section 12.03.

5.5 Oxygen

(M. Liadova)

Oxygen sensors attached to the CTD rosette frames measured oxygen concentrations for the different CTD casts. In total 337 oxygen samples were collected from different depths of the water column to calibrate these sensors. There were two different CTD types: The stainless steel CTD (SS CTD) and the trace metal clean CTD (TM CTD). For SS CTD, there were two casts per station: deep and shallow. For the SS CTD, 5 samples were collected in the deep casts, and 1 sample in the shallow cast. For the TM CTD, 6 samples were collected per cast. For Station 24, 7 samples were collected from deep cast of SS CTD and from TM CTD. The samples were collected bubble free and as soon as possible after the CTD was on deck. 6 different depths were chosen the same pattern: The highest and lowest depths, the oxygen minimum zone and depths that showed no or the weakest possible gradient. For the TM CTD, the samples were taken inside the clean lab and fixed with the fixating reagents outside of the trace-metal-clean lab to reduce the chances of contamination of other parameters being sampled. Oxygen concentration samples were analysed during the cruise using the Winkler (1888) Method. Two TITRONIC universal Piston Burette machines were used: one for sodium thiosulfate titration and one for iodate standardization. 17 oxygen samples were taken to calibrate EXO oxygen sensors. 4 oxygen samples were taken from the sediment water after the incubation experiments. See Sections 12.02-12.07.

5.6 Macronutrients

(A. Mutzberg)

The distribution of nutrients in seawater is a key for understanding the biogeochemical processes, and their signatures allow the differentiation between the various water masses in the ocean. In addition, they are used to identify leaking bottles due to their well-defined and oceanographically consistent distributions. Every Niskin bottle fired from every single cast depth was sampled for nutrient analysis onboard. Seawater was collected in 15 mL polypropylene sample vials. Containers and caps were rinsed three times with the water of the sample before the actual sampling. Samples were placed immediately in the fridge after collection (4°C in darkness) in case they could not be immediately analyzed.

Analysis of macro nutrients was undertaken on board by segmented flow injection analysis using a QUAATRO39 (Seal Analytical) auto-analyzer including a XY2-autosampler unit. Nano molar nutrient samples were frozen at -20 °C and will be shipped back to GEOMAR for analysis. The system set-up included 4 channels for nitrate + nitrite (TON), silicate, nitrite, and phosphate. The analytical methods followed during the cruise correspond to those described by QuAAtro Applications: Method No. Q-068-05 Rev. 11 for TON, Q-066-05 Rev. 5 for Silicate, Q-070-05 Rev. 6 for Nitrite and Q-064-05 Rev. 8 for Phosphate. A total of ~2430 macro molar nutrient samples collected for all Ti-CTD-casts, stainless steel-deep and shallow-CTD-casts were taken and subsequently measured on-board. Around 1000 nutrient samples were additionally taken from the stainless steel CTD casts to be shipped back at -20°C for

Nanomolar nutrient measurements at GEOMAR on a modified nano set up QUAATRO39 (Seal Analytical) auto-analyzer. Additionally, ~370 samples for dissolved organic nitrogen and phosphorus (DON / DOP) collected by all Meteor stainless steel-CTD-casts and were also measured after digestion on board. The digestion was done by using 40 mL of seawater sample in a 50 mL Duran glass bottle by adding Oxidizing decomposition reagent, Oxisolv ® Merck. The digestion happened in an overpressure cooker at 121 \degree C for 90 minutes. Ca. 50 Tow fish samples from the near surface, with expected concentration values in lower nanomolar concentrations for Nitrate + Nitrite and Phosphate, were stored at -20°C to be analyzed at GEOMAR with the macro set up and modified nano set up QUAATRO39 (Seal Analytical) auto-analyzer. Ca. 380 Nutrient samples were taken from the Multicorer casts and were analyzed on board with the macro set up QUAATRO39 (Seal Analytical) auto-analyzer. Ca. 250 Nutrient samples were taken by the Aerosol-collector to be analyzed on board with the macro set up QUAATRO39 (Seal Analytical) auto-analyzer. Finally, 4 rainwater nutrient samples were taken to be analyzed on board with the macro set up QUAATRO39 (Seal Analytical) auto-analyzer.

Certified Reference Material for Nutrients in Seawater (RMNS) was used in every run, in order to I) ensure repeatability and reproducibility between analytical runs and to II) validate the accuracy of concentrations measured. Nutrient analysis was validated with KANSO CRM, Lot-No. CL for macromolar nutrients.

5.7 Ammonium

(E. Achterberg)

Samples (15 mL) for ammonium were taken from Niskin bottles deployed on the SS-CTD down to ca. 300 m depth. Samples were immediately frozen at –20 °C and will be analyzed upon return to GEOMAR. Analysis will be conducted on thawed samples using the OPA method: OPA reagent will be added followed by a 24 h incubation time, during which the samples will be kept in the dark at room temperature with caps tightly closed. Subsequent detection of ammonium will be performed on a Carey Eclipse fluorimeter. Calibration will be carried out using standard additions with standards prepared using low ammonium concentration deep ocean waters (> 600 m).

5.8 Helium

(E. Achterberg)

³Helium is a conservative tracer for hydrothermal vent inputs and were sampled at some selected stations. Where sampled, helium isotopes were sampled first from the Niskins of the SS-CTD. Samples were collected in a copper pipes, which were connected to the Niskin bottle via plastic tubing, with water flowing until bubbles in the tube were removed. While the water was flowing, the pipe was closed using an electrical drill and a ratchet. Helium isotopes will be analysed at the University of Bremen.

5.9 Trace Metal Sampling

(D. Jasinski, Y. Ruan and A. Nicolas)

Trace metals (TM) play a key role in marine ecosystems, as they are essential micronutrients and can also be toxic. The Eastern Mediterranean Sea is characterized by a complex water masses circulation, combined with small-scale features and is strongly impacted by anthropogenic climate change and pollutions. Measuring the distribution of trace elements in the water column will help understanding the complexes sources, sink, fluxes, transport mechanisms, natural and anthropogenic induced processes taking place in the area. To limit contamination of the samples, acid-cleaned *Niskin* bottles were deployed on a Titanium rosette, equipped with a CTD. Samples were collected inside the "clean lab container" over pressurized with HEPA-filtered air, acidifications and filter handling was done inside a laminar flow hood. *Niskin* bottles were kept in the clean lab between casts. **Oxygen**: Oxygen samples were collected from few *Niskin* bottles, always sampled first and analysed on board (See Oxygen section). **Salinity**: Salinity samples were collected from several *Niskin* bottles per cast (See Salinity section). **Nutrients**: Nutrients samples were collected from all *Niskin* bottles and analyzed on board (See Macronutrients section). **THg**: Unfiltered total Mercury samples were collected at every station from 12 depths, 60 mL acid cleaned glass vials were rinsed 3 times before collection (overflow). Samples were acidified with 300 μL (*Optima* or TM grade) HCl (final concentration 0.5%). **MeHg**: Unfiltered total Mercury samples were collected at every station from 12 depths, 125 mL plastic bottles were rinsed 3 times before collection (overflow). Samples were acidified with 620 μL (*Optima* or TM grade) HCl (final concentration 0.5%). **DOM/TM**: Water samples were collected in 2 L bottles (rinsed 3 times with sample before collection) from few depths, samples processed on board (cf Organic matter sampling and analysis section). **Dissolved TM**: Filtered water samples (using Pall *Acropack* filters 0.2 μm) were collected into acid cleaned (following GEOTRACES protocols) 125 mL Nalgene LDPE bottles (rinsed 3 times with sample before collection). Samples were acidified with 175 μL *Optima* grade HCl (Fisher Scientific). **Particulate TM**: 0.2 μm, 43 mm diameter PES filters were cut from larger filters using ceramic scissors and then soaked in 0.1 M HCl acid (for some time in analytical grade and then for some time in *Optima* grade acid). Seawater was passed through the filters whilst under pressure from filtered N_2 gas (0.5 bar). The volume of seawater filtered through each filter was measured. Acid-cleaned plastic filter holders were used on the first station and then rinsed with MQ in between samples/stations. Blank filters were collected several times during the cruise. All samples were stored at -20°C. **Major elements**: Filtered water samples (using Pall *Acropack* filters 0.2 μm) were collected into acid clean 5 mL vials. (See Major Elements section). **Soluble TM**: 12 filtered water samples per cast (using Pall *Acropack* filters 0.2 μm) were collected into acid washed (following GEOTRACES protocols) 125 mL Nalgene LDPE bottles (rinsed 3 times with sample before collection). These bottles were reused between stations – they were placed in acid bath overnight and rinsed with Milli-Q. Samples were then "ultrafiltered" with 0.02 μm filters (Anotop, 25 mm diameter) using a peristaltic pump. Filters were used for 3 different samples, flushed with 0.1M HCl, Milli-Q and then sample before collection into 30 or 60 mL acid washed (following GEOTRACES protocols) Nalgene LDPE bottles. Then acidified with 40 or 80 μL of *Optima* grade HCl (Fisher

Scientific). **Zn isotopes**: 1L LDPE bottles were rinsed 3 times with filtered (using Pall *Acropack* filters 0.2 μm) seawater sample before collection. **Dissolved Nd/Pb/Sr isotopes**: "Cubitainers" of various sizes (ca. 1-10L) were rinsed and filled with filtered sample (through the 0.2 μm PES filters used for particulate sample collection). **Particulate Nd/Pb/Sr isotopes**: The same procedure as for particulate TM.

5.10 Major Elements

(Y. Han, Z. Steiner)

Major elements are mostly conservative in seawater and have high concentrations and long residence times in the ocean. However, major dissolved cations participate in most biological and geochemical reactions and thus variability in their concentrations outside the variability determined by changes in salinity informs of a multitude of reactions and can be used to quantify key processes in the marine environment. As seawater Mg:Ca and Sr:Ca ratios are essential biogeochemical parameters, their concentrations reflect the dynamic exchange of important elements between the solid Earth, ocean water masses, the atmosphere, and processes such as biological precipitation of inorganic minerals and *post-mortem* dissolution. On this cruise, the investigated processes that may govern distribution of the cations Ca, Mg, Sr, Ba and Li are marine currents and circulation, biogenic inorganic mineral precipitation and dissolution, atmospheric inputs, and deep sedimentary sources that reach the surface as mud volcanoes and brines.

Samples were collected from all stations visited during the cruise and at all depths to quantify the distribution of major elements, identify the processes that govern variability in the distribution of the major elements and evaluate their budgets in the Eastern Mediterranean basin. The samples were filtered using 0.8/0.2 μm filter cartridge (AcroPak 500, Pall) from the trace metal CTD. Filtered underway samples were collected from the towfish system regularly. Samples were acidified with HCl (Optima grade; Fisher Scientific) and stored at room temperature. The ratios of the major dissolved cations Ca, Mg, Sr, Ba and Li to Na will be analyzed at GEOMAR using a Varian-720 ICP-OES (Inductively Coupled Plasma Optical Emission Spectroscopy) (Steiner et al., 2020).

5.11 Carbonate System

(L. Qiu)

To assess how climate change affects carbonate system in the eastern Mediterranean Sea, total alkalinity (TA), partial pressure of $CO₂$ ($pCO₂$), pH and dissolved oxygen (DO) in surface water were observed using an *in situ* TA analyzer (ISA−TA) with about 7-min sampling interval, a *p*CO2 sensor (HydroC, 4H−Jena engineering GmbH, Germany) with 10 s sampling interval, a pH sensor (sunburst sensor, USA) with 15-min sampling interval and an EXO1 probe/sensor with 1 min sampling interval, respectively (Table 5.01). A suite of sensors (including ISA-TA, pCO2, pH, EXO1) were placed in a 68 L tank on the sink of 'Geo' lab, which was continuously supplied with surface water (from 2.5 m depth) from the underway water supply during observations. The flow rate was about 30 L/min allowing for fast exchange of the entire water volume inside the tank. Although the fast water exchange, the turnover time of two minutes should be considered when processing the data.

Underway samples for dissolved inorganic carbon (DIC) and TA were collected once daily (around 30 samples) and were poisoned with mercuric chloride $(HgCl₂)$ prior to analysis on land. Profile DIC and TA samples (around 700 samples) collected from stainless steel CTD were immediately measured using ISA-TA in the ship. Underway discrete samples for TA will be analysed following the Gran titration method (Gran 1952) using a semi−automatic potentiometric titration system (AS−ALK2, Apollo SciTech Inc., USA) (Cai, Hu et al. 2010). Discrete samples for DIC will be measured using an Apollo DIC Analyzer (Model AS−C3).

Preliminary results from profiling observations indicate that TA values are higher at the surface than at the bottom in the eastern Mediterranean Sea (Fig. 5.01).

5.12 Dissolved Organic Carbon (DOC)

(E. Achterberg)

DOC samples were taken and acidified to pH 2 in the lab. Shallow samples from ca. 300 m upward were filtered through an ashed GF/F filter. DOC samples will be analysed at GEOMAR using a Shimadzu TOC/TDN instrument.

5.13 In Situ Pump Deployments

(J. Blanke, E. Achterberg)

Sinking of particulate organic matter (POM) links $CO₂$ fixed through photosynthesis by phytoplankton in the surface ocean with carbon storage in the ocean interior. This pathway has been conceptualized as the biological carbon pump (BCP), and represents a key carbon sequestration mechanism. However, the bioavailability and bacterial transformation of POM, which are key determinants of BCP efficiency, remain poorly understood. Biomarkers (e.g., amino acids and amino sugars) provide powerful tools for insight into the bioavailability and transformations of POM.

Particles were collected using in-situ pumps. Two, sequentially filtered size fractions (51 and then 1 μm) were obtained by PETEX meshes at each depth, with a standard pump time of 2.5 hours. Nine pumps were used in total, including six challenger-oceanics from National Oceanography Centre Southampton (UK) using 293 mm diameter PETEX meshes and pumping 1800-3200 L, two Kiel In situ Pumps (KISP) pumps, one equipped with a 293 mm diameter PETEX mesh, the other with a 142 mm diameter size mesh and one McLane pump, equipped with 142 mm diameter size meshes. The KISP pumps were equipped with only the 1 μm mesh, while the McLane pump had both pore-sizes equipped. Both of these smaller pumps pumped around 300-500 L. In alternation, the particles on the mesh were either rinsed using filtered seawater from 1000 m depth into beakers and then divided into four parts (POC, amino acids, amino sugars) or using thorium-free underway seawater and divided into five parts (POC, POP, PIC, BSi and ²³⁴Th) with Folsom splitter for 51 μ m and 1 μ m, respectively. Additionally, a quarter was cut from the filter and frozen at -80°C on days thorium was sampled. At stations 4, 6, 8, 10, 12, 17, 19, 21, 23, 25, 27 and 29 nine depths between 20-1000 m were sampled, while at stations 1, 3, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26 and 28 nine depths between 20-700 m were sampled. The partitioned seawater for POC, POP, amino acids and amino sugars were filtered onto pre-combusted GF/F (Fisherbrand; Product number: 11750483) filters and then stored at -20°C, seawater for BSi and PIC were filtered onto 0.4 µm polycarbonate membrane filters (cytiva; Product number: 10417706) and onto QMA-filters (Whatman; Product number: 1851-025) for 234Th. At Stations 6, 8, 10, 12, 17, 19, 21, 25, 27 a quarter was cut from every filter and dried at 55 °C for Nd and Sr isotopes. These filter-cuts will be analyzed by Adi Torfsteine at the Hebrew University in Jerusalem.

Samples for POC, PN and bulk $\delta^{13}C$ and $\delta^{15}N$ determination will be fumed with concentrated hydrochloric acid (HCl) to remove inorganic carbonate followed by oven-drying at 60°C for 24 h, and analyzed using a Flash EA IsoLink CN elemental analyzer coupled with a MAT 253 plus IRMS (Thermo Fisher Scientific, Germany).

Pretreatment of AS samples will follow the method of Zhu et al. (2014). Briefly, GF/F filters or ground sediments will be hydrolyzed using 6 M HCl at 105°C for 8 h. The hydrolysates will then be neutralized with 6 M potassium hydroxide (KOH) to a pH ~ 6.8 and centrifuged immediately. The supernatant will be taken through solid phase extraction (SPE) cartridges to remove salts, and then eluted with methanol and dichloromethane. The eluent containing AS will be concentrated under nitrogen and then redissolved in Milli-Q water for concentration and stable carbon isotope analysis. Individual AS concentrations will be quantified using an ion chromatograph (IC, Dionex ICS-5000+ SP) coupled with an electrochemical detector.

Concentrations of D- and L-amino acids will be hydrolyzed using 6 M hydrochloric acid and then separated as *o*-phthaldialdehyde derivatives using a Thermo Fisher Scientific U3000 ultrahigh performance liquid chromatography (UPLC) system equipped with a Poroshell 120 EC-C18 column (4.6×100 mm, 2.7 μm particles). Further details of amino acid hydrolysis and chromatographic separation are provided by Shen et al. (2017).

5.14 Thorium

(J. Blanke)

²³⁴Th is the daughter isotope of the ubiquitous radiogenic ²³⁸U. The resulting disequilibrium between ²³⁴Th and ²³⁸U as the surface active ²³⁴Th gets scavenged by sinking particles can be used to calculate particle fluxes such as export of POC to deeper depths, an important aspect of quantifying organic carbon export rates in open ocean waters (Buesseler et al., 2006, Chen et al., 2008).

Sampling of dissolved ²³⁴Th and ²³⁸U occurred every other day at the Stations 1, 3, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26 and 28. Fifteen 4 L samples of unfiltered seawater were collected from the stainless steel CTD that was deployed with the in situ pumps. Samples were collected at 5-700 m depth for analyses of 234Th activity to calculate POC flux. At Station 19 two deepwater samples at 1000 m were taken as reference samples. All ²³⁴Th samples were acidified to pH <1.5 (~1.3) concentrated nitric acid (68 %) directly after sampling. Further, 100 μl of yield standard solution was added. After 8 h, the pH was neutralized to pH 8.2-8.5 (\sim 8.3) using concentrated ammonium solution (\geq 25 %). 50 μL of KMnO₄-solution and 50 μl of MnCl₂solution were added to co-precipitate 234 Th with MnO₂. After waiting for 8 h, the precipitate was filtered onto QMA filters, rinsed with pH 9 MQ water and dried at 50 \degree C for \sim 5h. Sampling bottles were cleaned with 500 mL of H₂O₂/HNO₃-solution and rinsed three times with MQ. Before sampling, the bottles were rinsed three times with seawater and the vials three times with filtered seawater from the according Niskin-bottle (for details on filters used for subsamples refer to the 'In situ pumps' Section)

At each Station where sampling of dissolved ²³⁴Th and ²³⁸U occurred, twelve samples for particulate 234Th, biogenic silicate (BSi), particulate inorganic carbon (PIC) and genomics were collected from the in situ pumps at six depth 20-700 m at the Stations 3, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26 and 28. Here the two pore-sizes, 1 µm and 51 µm, were sampled for each of the six depth accounting for two samples per depth. The polyethylene meshes were rinsed with thorium-free underway seawater (pH brought up to 8.2-8.5 and 125 μL of KMnO4-solution and 125 μl of MnCl₂-solution were added to co-precipitate ²³⁴Th with MnO₂) and filtered onto a QMA filter for the particulate 234 Th and 0.4 μ m polycarbonate membrane filters for PIC and BSi. QMA-Filters were dried at 50 °C for \sim 5 h and the membrane filters for \sim 5 h. After drying, each QMA filter was covered in plastic foil and wrapped with aluminum foil onto the sample holder. Membrane filters were stored at room temperature in petri dishes for further analysis at GEOMAR.

For all samples of dissolved and particulate ²³⁴Th, total beta activity was measured using a Risø low-level beta GM multicounter until the error of the measurement reached values $\leq \pm 5$ % ($\sim \pm 2.5$ % most often). Measurement time ranged from ~ 8 h for the particulate ²³⁴Th samples to \sim 4 h for the dissolved ²³⁴Th samples. After the beta counting, the samples were stored at room temperature. All 234Th samples will be measured after 6 months for baseline radioactivity correction. ICP-MS analysis of 230Th will be conducted to determine the individual yield for Thorium recovery rates. From every depth of 234Th sampling, 10 mL of filtered seawater were sampled for analysis of ²³⁸U. Samples were collected in 15 mL screw-lid vials, acidified with 20 μL of concentrated HCl (32%) and stored at room temperature. The concentration of 238 U will be determined at GEOMAR using ICP-MS, PIC concentrations and δ13C will be determined at Woods Hole Oceanographic Institution and BSi will be determined at GEOMAR using the Mortlock and Froelich (1989) method.

5.15 Microplastics Sampling

(L. L. Cai)

The aim is to further unveil the vertical profile of microplastics on the eastern sector of the Mediterranean Sea. Seawater and sediment samples were collected for quantification and characterization of microplastics. Seawater was retrieved from Niskin bottles that sampled at the bottom, middle and surface of the water column (the exact depths of bottom and middle were dependent on station depth and surface on weather conditions). In total 28 stations were sampled for microplastics in seawater. Dedicated 10 L Niskin bottles were used to ensure retention of all potential microplastic particles, as different plastic polymers and shapes express flotation discrepancies. The seawater was then filtered through mixed cellulose ester filter papers (47 mm diameter, 0.45 μm pore size) by using a glass filtration system (Figure 5.02). Filter papers were then oven-dried at 55 °C for 1 hour before stored individual in plastic petri dishes for transportation to CMMI laboratories. There, the filters will be made transparent using immersion oil and examined under a light microscope for microplastic quantification and characterisation.

Fig. 5.02 Filtration system for water samples and petri dishes with dried filter papers ready for storage.

Seafloor sediment was sampled using a multi-corer system, from which the top 5 cm sediment of a single core was utilized for microplastics analyses (Figure 5.03). In total 19 stations were sampled for microplastics in sediment. Sediment was collected using a metallic spatula and placed in a glass jar. Samples were then oven-dried for \sim 72 h at 55 °C. Analysis on the sediment samples will take place at CMMI, where sediment will be weighed, and microplastics will be extracted using a density separation method based on a hyper-saline solution. During this process, the supernatant will be retrieved and filtered using a glass filtration system onto a mixed cellulose ester filter paper (47 mm diameter, 0.45 μm pore size). Microplastic quantification and characterization from filters will be carried out via microscopy. All the analyses will involve standardised methods to minimize microplastic contamination.

Fig. 5.03 A: multi-corer after a successful deployment. B: extraction of corer for sampling. C: microplastics sample collected.

5.16 Molecular/Elemental Signatures of Particulate and Dissolved Organic Matter

(J. Li)

To assess the association between particulate and dissolved organic matter (POM/DOM) and metals, I collect both the POM and DOM samples using a setup in a cold lab. Two-liter unfiltered seawater from 6 depths between 20 m to 1000 m (including DCM layer) collected in TM CTD of 25 stations was used to concentrate DOM and POM (Section 12.01). In total of 147 dissolved and particulate organic matter samples were collected onto ENV solid phase extraction cartridges (500 mg) and Sterivex PVDF 0.22 µm filters, respectively. The cartridges for dissolved part were frozen at -20°C and will be extracted and analysed by high performance liquid chromatography – electrospray ionisation mass spectrometry – inductively couple plasma mass spectrometry (HPLC-ESI/MS-ICP/MS). The filters for particle part were frozen at -80°C and the organic matter will be extracted from particles with a mild extracting agent that solubilises organic matter with minimal molecular alteration at pH 7.8 and analysed by high performance size exclusion chromatography (HPSEC) -ESI/MS-ICP/MS. We will quantify Sulfur, Phosphorus and metals as a function of molecular size and hydrophobicity (Gledhill et al., 2022). Based on this, the relationships between elements in organic matter and determine the association between elements and size fractions will be examined. Our

overarching aim will be to examine changes in these relationships within the Mediterranean Sea and relate them to the biogeochemical regime in the region.

5.17 Alkaline Phosphatase Activity and Particulate Organic Phosphorus

(J. Li)

AP is a typical phosphorus limitation bioindicator in the oligotrophic sea area. To investigate the AP activity profile of Mediterranean Sea, 10 mL seawater samples from 9 depths between 6 m to 250 m collected in SS_CTD of 30 stations were collected to carry out bioassay (Section 12.11). Methylumbelliferyl phosphate (MUF-P; 50 nmol L-1) was used as the organic phosphate substrate and directly following the protocol of Browning et al. (2017). Fluorescence was measured on a plate reader (FLX800TBI, BioTek) with Gen 5 software using an excitation wavelength of 365 nm and an emission wavelength of 455 nm. Following MUF-P spiking, fluorescence measurements were performed at 0 h and 3 h. AP activity indicated by MUF-P turnover time (h^{-1}) was calculated as fluorescence of 50 nmol L^{-1} 4-methylumbelliferone (MUF) divided by the initial (0 h to 3 h) slope of fluorescence time course (fluorescence per hour). Meanwhile, 8 L seawater samples from the same depths were collected and filtered onto the pre-combusted (combusted in a Muffle Furnace at 450°C over 5 h) 25-mm GF/F membranes and put in the freezer. Before analysis, each membrane will be autoclaved at 121°C for 30 min with 5% acid potassium persulfate $(K_2S_2O_8)$ and the DIP concentration in the solution will be determined by a phosphorus molybdenum blue spectrophotometry.

5.18 DOP Incubation Experiment

(J. Li)

To investigate the bioavailability of one typical terrigenous DOP species, phytic acid (PA), we carried out 4 microcosm incubation experiment. Seawater samples were collected from towfish pump (depth of 5 m). Samples were then transferred into 4.5-liter transparent plastic bottles and incubated in a water tank on deck with a continuous flow of surface seawater with blue lagoon filter (Lee filters) to adjust light to ambient irradiance (Section 12.12). Four groups were setup: Control group (no nutrient added), PA group (0.5 uM phytate added), N group (2 uM ammonia added), and PA+N group (0.5 uM phytate and 2 uM ammonia added), each in triplicate. After 2 days and 5 days incubation, cells were collected for Flow cytometry, chlorophyll (GF/F filter), AP activity, and DNA (47 mm 0.22 um PC membrane). The filter samples will be used for RNA and DNA extraction, and the community structure changes and differential gene expression of phytoplankton under PA addition will be analysed.

5.19 Rainwater Collection

(A. Nicolas)

Rain water was collected using acid cleaned plastic funnel and 1L LDPE bottles. Different kinds of sub-samples were collected depending on the volume of rain collected and processed as for seawater samples (See 'Macronutrient' and 'Trace Metal' Sections).

5.20 HPLC, POC/N, Chlorophyll-a

(A. Blachinsky)

Samples were collected to characterize phytoplankton biomass, and community structure throughout the cruise transect. Samples were collected at 28 stations at 8 depths throughout the upper water column (7-200 m for chlorophyll a); and 6 depths throughout the upper water column (7-120 m for HPLC and POC/N). Depths of sampling were selected after consultation with the fluorescence trace on the CTD. Water was collected in 10L opaque carboys and processed immediately after collection. Samples were collected for analysis of the following: - Chlorophyll-a concentrations: 500 mL samples were filtered onto Fisher MF-300 GFF filter pads and extracted for 20-24 hours in 10 mL 90% acetone in a -20 °C freezer in the dark before measurement on a Turner Designs trilogy fluorometer. High-Performance Liquid Chromatography (HPLC): 3-4 L seawater was filtered onto Fisher MF-300 GFF filter pads and placed directly into a -80 °C freezer. These will be analysed on return to GEOMAR. Particulate Organic Carbon (POC): 4L seawater was filtered onto pre-combusted (4h; 450°C) Whatman glass microfiber filters, Grade GF/F. To remove inorganic carbonate, POC samples were dried overnight in an oven at 60°C for 12-14 hours. These will be analysed on return to GEOMAR.

Figure 5.04 Spatial changes over the course of the M197 cruise in the extracted chlorophyll *a* in units of μ g/L calculated by a Turner Designs trilogy fluorometer. ODV interpolation (x - 18, y - 20) Data have not gone through quality check.

5.21 Microbial Genomics

(C. Eckmann, J. Ivaldi)

During this cruise we conducted sampling to characterize the microbial community throughout the water column. We collected water for DNA, flow cytometry (FCM), and meta-genomic (meta-g) samples. These samples will be further processed in the laboratory of Alexandra Worden at the Marine Biological Laboratory in Woods Hole, Massachusetts by C. Eckmann and Rachele Spezzano later this year.

For DNA samples, the V1V2 region of the 16S rRNA gene (present in bacterial DNA and in the bacteria-derived plastid of eukaryotic phytoplankton) will be amplified to allow for the identification and calculation of relative abundance of microbes. FCM samples will be analyzed to quantify absolute abundance of microbes identified based on size and fluorescence to a broad taxonomic and/or trophic category (e.g., cyanobacteria, heterotrophic bacteria, picoeukaryotes). Together, these will allow for precise quantification at a fine taxonomic resolution for many important marine microbes. Meta-g samples will complement this information by providing insights into the metabolic potential of this microbial community. DNA samples were collected in duplicate from 6 depths in the photic zone, including the deep chlorophyll maximum (DCM) when present, and 6 depths from below the photic zone to the seafloor. 1 L of seawater per replicate was filtered using 47 mm diameter 0.2 µm pore size filters. FCM samples were collected from the same depths but in triplicate to allow for differences in flow cytometry settings to capture a broad range of particle sizes. 1 mL of seawater was fixed using 1 μ L of 25% glutaraldehyde, incubated in the dark for 20 minutes, then flash-frozen in liquid nitrogen. Meta-g samples were collected by filtering approximately 20 L of seawater from the surface (5-7 m) or the DCM through a 20 µm mesh onto 142 mm diameter 3 µm filter then sequentially onto a 142 mm diameter 0.2 µm filter. For meta-g samples, corresponding size-fractioned DNA samples were also taken. All samples were stored at -80°C. The total number of samples was 720 DNA, 1,080 FCM, 52meta-g, and 52 sizefractioned DNA.

Subsets of the in-situ pump filters were collected on days when Jana Blanke sampled for thorium. These will be processed along with the water column DNA samples to determine the microbial community contributing to the particulate fractions (51 μ m and 1 μ m pore size). In addition, we collected samples for lipid analysis using the same methods as for the DNA samples, except using a PVDF membrane filter and sampling every other day from 3 depths: surface (5-7m), DCM, and 300 m.

5.22 Primary Productivity, Dark Carbon Fixation and Nitrogen Fixation

(T. Reich)

Inorganic carbon assimilation by phytoplankton is fundamental to life in the ocean and greatly influences the oceanic carbon cycle. These primary producers are at the base of the oceanic food web and are the first to convert dissolved $CO₂$ into available organic carbon for the higher trophic levels. Measuring primary production (PP) rates at the eastern Mediterranean's euphotic zone provides vital information on photosynthetic activity by photoautotrophs within an ultra-oligotrophic sea. These rates facilitate our understanding of the potential for carbon

export to the deeper oceanic layers. In addition to measuring photosynthetic carbon fixation and export it is important to measure the inorganic carbon that gets fixed by chemoautotrophs (i.e. Dark carbon fixation (DCF)) within the photic zone and also deeper in the aphotic zone. Combining PP and DCF provides a better assessment of new organic carbon production and the potential of carbon export to depth.

Dinitrogen fixation $(N_2$ fixation) by a subset of prokaryotic phytoplankton, bacteria, and archaea, is a major source of new nitrogen that is added to the world ocean's – especially in the nitrogen limited surface waters. Historical evidence, based mostly on isotopic composition of $15N/14N$, indicates that N₂ fixation contributed significantly to organic matter in the Mediterranean. Overall results from \sim 15 years show that the potential for N₂ fixation exists and a diversity of diazotrophs are recorded across the Mediterranean. Yet, in the eastern Mediterranean Sea actual volumetric and areal rates of N_2 fixation are generally very low (range: 0 to 17 nmol N L⁻¹ d⁻¹ and 0-50 µmol N m⁻² d⁻¹ respectively in the upper water column) and no typical seasonal patterns were observed. More recent work shows N_2 fixation also occurs in the dark below the photic zone and has also been measured in the Mediterranean. Here we targeted several representative stations and depths where diazotrophs (N_2) fixing organisms) may be found and performed analyses to determine N_2 fixation rates.

Sampling information:

Euphotic zone sampling - Samples were collected from 6 depths at most stations along a transect from the east to west sides of the levant basin of the Mediterranean Sea. 7 Samples of 1.2 L were collected using a SS-CTD at 6 depths with some variations (7, 20, 40, 80, 100-DCM and 120 meters). 3 bottles were placed in light adjusted incubators to match source levels (100%, 50%, 25%, 10%, 1% 0.1% PAR of total radiation) for total production rate measurements and 3 in dark incubators to account for the chemosynthetic activity.

Aphotic zone sampling - Samples were collected from 6 depths at most stations along a transect from the east to west sides of the levant basin of the Mediterranean Sea. 4 Samples of 1.2 L were collected using a SS-CTD at six depths (a changing bottom depth, 800, 600, 400, 200, 150 meters). Samples were incubated for 24 hours prior to filtration in dark incubators at ambient sea water temperatures (flow through to the ship).

Nitrogen fixation – At stations 6, 11, 17 and 25 water was collected from 3 depths (15m, DCM and 120m) in triplicate 4.6 L bottles using a SS-CTD.

Bioassay aggregation experiment - The objective of these bioassays was to evaluate whether particulate organic carbon facilitates DCF rates and can elevate the potential export of newly fixed carbon from the euphotic zone into the deep ocean. 4 bioassays were set up during the cruise. Each assay included triplicate 4.6 litter bottles of 2 sets of samples (control and treated) from 2 depths (DCM and below with variation according to the station profiles). To the treated samples Gum Xanthan was added to give an organic carbon source in particulate form. All samples were spiked with and placed in dark incubators for 24 hours. Before and after 24 hours all treatments were sampled for 13C uptake, FCM, TOC, TEP and DNA.

• Brief experimental protocols **Primary productivity and DCF** - Samples were spiked with NaH¹³CO₃ (99 atom% 13C, Cambridge Isotope Laboratories) final concentration 20 μ M stock. Bottles where placed in light adjusted on-deck incubators to match source levels for total production rate measurements or dark incubators to account for the chemosynthetic activity with running surface seawater. After incubation samples were filtered in precombusted GF/F (Whatman, 0.7μ m pore size, 25mm) filters and placed over night in a 60 C oven for drying.

• **Nitrogen fixation** - Each bottle was spiked with 40ml ¹⁵N₂ stock (98 atom%, Cambridge Isotope Laboratories) dissolution method (final concentration is TBC) and 460µl NaH¹³CO₃ (99 atom% 13C, Cambridge Isotope Laboratories) final concentration 20 μ M. Bottles were placed in incubators with neutral-density mess or complete dark to adjust to light levels of sample source and incubated for 24 hours in an on-deck incubators with running surface seawater. After incubation samples were filtered in pre-combusted GF/F (Whatman, 0.7μ m pore size, 25mm) filters and placed over night in a 60 °C oven for drying.

Future analyses – All filtered samples are going to be sent to mass spectrometry analysis to determine 13C and 15N assimilation rate in each sample at the end of the cruise and are expected to take 3 months to have results. FCM, TEP, TOC and DNA samples are going to be analyzed and processed at IOLR in the weeks after the cruise and are expected to be ready after 2 months. All samples will be processed by or under the supervision of T. Reich

5.23 Phytoplankton Photo-physiology

(I. Berman-Frank)

Single-turnover active fluorometry (STAF) is a technique used to assess phytoplankton photophotophysiology and estimate photosynthetic rates. During M197, A Chelsea Technology Ltd LabSTAF instrument (More information about the instrument and approach can be found in the instrument handbook, available on the Ocean Best Practice Repository: https://repository.oceanbestpractices.org/handle/11329/1531.4.rs) was utilized to examine several objectives:

- 1) What are photophysiological changes along the cruise track (surface waters)?
- 2) What are the diel photophysiological signatures and do these change in different mesoscale features (cyclonic versus anticyclonic eddies)
- 3) How do photophysiological signatures vary with depth in the photic zone?
- 4) What do the photophysiological signatures reveal about community composition of the surface phytoplankton?
- 5) How do nutrient and aerosol enrichments impact the photophysiological response?

Data sets were collected for underway surface populations. The data was collected continuously to examine daily patterns at each station (diel changes in photophysiological activity) and also between stations along the cruise track within the allowed work-areas. Additionally, data was collected daily at each station for 6 pre-determined discrete depths within the photic zone from the shallow CTD-Rosette. These discrete samples were run using a measurement protocol with 5-6 light levels to obtain photosynthetic parameters for the dark regulated state and also be able to derive gross oxygen evolution of PSII. For the different depths photosynthetic excitation profiles (PEP) were measured using 7 different excitation wavelengths. These measurements allow for automated spectral correction of derived

photosynthetic rates and provide a way to track changes in phytoplankton species composition. Further, for all samples dual waveband measurements (DWM) were acquired at two emission wavelengths. DWM values are being tested to provide a proxy for the so-called pigment packaging effect.

The LabSTAF was connected to the continuous seawater supply of the vessel throughout the cruise (membrane pump) and was run using a measurement protocol that included 8 light levels (Auto adjusted) to obtain fluorescent light curves (FLCs) that allows the derivation of the dark-regulated photophysiological parameters and estimates of photosynthetic rates at 7 reference light level. The limited biomass in this region (and especially in the eastern parts of the transect required 80 sequences/acquisition and only one ST flash. On 27 Jan, the peristaltic pump that exchanges the water in the sample chamber broke down and "continuous" measurements were done manually when possible, at a frequency of one FLC per half hour (using the same protocol as above). See Section 12.13 for a summary of measurements and protocols.

The LabSTAF was additionally utilized in all bioassay experiments done on board M197 (T. Browning, A. Blachinsky, J. Li). These bioassays were set up to test what the limiting nutrient/s are for the ambient phytoplankton populations. Nutrient additions of varying N and P sources (both inorganic and organic) were added and phytoplankton populations incubated for 48 h and 5 days.

The LabStaf was also used to measure changes in phytoplankton population and activity for bioassays testing the impact of aerosols and leached nutrients from aerosols (For B. Herut and B. Ankri).

Figure 5.05 Spatial changes over the course of the M197 cruise in the photosynthetic efficiency of photosystem II (Fv/Fm) and in the derived gross oxygen release by PSII (GOPII) – in units of mmol O_2 m⁻³ h⁻¹ calculated from the LabStaf basic parameters for the underway surface water – 5m depth.

5.24 Nitrogen Uptake Rates and Urea Concentration Analysis

(A. Blachinsky)

Uptake experiments were carried out using ¹⁵N urea, ¹⁵N nitrate, and ¹⁵N ammonia + ¹³C-HCO₃ as tracers. To carry out the uptake experiments as close to in-situ conditions as possible, we planned to add the labelled tracer at \sim 10% of the ambient concentration, and no more than 20%. To assess the desired concentration, uptake rates experiments were conducted following the nitrate concentrations examined daily on RV METEOR. However, it was always necessary to make the addition in the field before we had determined the actual ambient concentrations. In the case of nitrate – we used the measured data from samples collected the previous day to estimate the amount of tracer to add. Based on these considerations, ammonium was estimated to be 0.02 -0.1 μ M and urea was estimated to be 0.1 -0.2 μ M. We used the ratio between ammonium to urea concentration in similar oligotrophic systems, in addition to the ammonium concentration from our previous measurements at the 800m station, to predict the urea concentrations. The actual percentage of the $NO₃$ tracer and the ammonium and urea addition are given in Section 12.14.

The water was sampled into 4.5L bottles from trace metal clean tow-fish from 5 m depth. This volume was chosen to avoid results below the detection of the mass spectrometer by adding 10% of ¹⁵N of assumed ambient concentration (\sim 1 nM ¹⁵NO₃). The incubation treatments were first spiked with the ¹⁵N and then incubated at ambient water temperature in flow-through water tanks on deck, with blue lagoon filter (Lee filters) to adjust light to ambient irradiance. The t_0 treatments were spiked and immediately filtered (3L). 3L of The remaining samples were filtered at t = 4-6 hours onto pre-combusted (450°C; 4h) 25mm GF – 75 (0.3 μ m retention

The filters were then stored in a -20° C freezer. The filters were dried overnight at 60°C oven before transport to Israel (Morris Kahn marine station). Filters will then fumed in a desiccator for 24 h in the presence of concentrated HCL and then redried at 60°C for 1 hour.

The samples will be sent to Prof. Mark Altabet to be analysed by Continuous Flow-Isotope Ratio Mass Spectrometer (CF-IRMS) at the MBL Stable Isotope Laboratory which is located at the Ecosystems Center on the campus of the Marine Biological Laboratory in Woods Hole, Massachusetts.

Urea concentrations from $t=0$ h samples will determine using a frozen filtered nutrient samples. The samples will be analyzed by an adaptation of the manual method using a 3 day incubation at room temperature. 35 ml of thawed sample will be placed into a 50 ml prewashed dark bottle (1 time 10% HCL follow by 3 times distilled water followed by 1-time *Milli-Q water)*. To each bottle, 2.5 ml of Reagent A (diacetyl monoxime together with thiosemicarbazone) and then 8 ml of Reagent B (ferric chloride solution) will be added. The samples will transfer immediately after the addition of reagents into a dark drawer at controlled room temperature (24°C) for 68-72 hours of incubation. The samples will subsequently be analyzed in a dark room using a GENESYS 150 UV-Vis Spectrophotometer with a 10 cm cuvette at 520nm.

5.25 Nutrient Addition Bioassay Experiments

(J. Li, A. Blachinsky, T. Browning, I. Berman-Frank)

A total of 4 microcosm nutrient enrichment experiments were carried out to assess for limiting nutrients to the phytoplankton community to (Section 12.15). The first two experiments were carried out with ammonium, phosphate, and dissolved organic phosphorous while the last two were carried out with the addition of urea and nitrate to the previous treatments. Treatments combinations are shown in Section 12.15. Seawater samples were collected from tow-fish pump (depth of 3-5m). Samples were then transferred into 4.5-liter transparent plastic bottles. Every treatment was conducted in triplicate. After spiking, the bottles were incubated in a water tank on deck with a continuous flow of surface seawater with blue lagoon filter (Lee filters) to adjust light to ambient irradiance. The treatment incubated on board for 5 days from the time of spiking $(t = 0)$. We used a PAR sensor to measure the incubation percentage from natural irradiance and at the end of the cruise.

Chlorophyll a, Single-turnover active fluorometry (STAF), and APA parameters are explained specifically in the relevant sections of this report.

Flow Cytometry to assess picophytoplankton and bacterial abundance: Two ml from each treatment was sampled at $t = 2.5$ days into a cryovial and fixed with a flow cytometry grade glutaraldehyde solution (0.125% final concentration) (NH_4^+ (as NH_4Cl) + PO₄ (as KH_2PO_4); NH₄⁺; PO₄; control; urea + PO₄; NO₃⁺ PO₄⁻)) or Paraformaldehyde solution (1% final concentration) (control; NH_4^+ ; DOP; NH_4^+ + DOP). Samples were incubated for 10 minutes in a dark box before flash freezing at liquid nitrogen. After the flash freezing, sampled were stored at -80°C. The FCM samples will be measured on a flow cytometer (FACSCantoII, BD) at the university of Haifa and then analyzed with FlowJo software (FlowJo v10.8.1).

5.26 Coccolithophores Abundance & d13C-DIC

(Y. Han, Z. Steiner)

Marine calcium carbonate production is an important process in modulating global ocean alkalinity, which impacts oceanic carbon dioxide release and absorption. Coccolithophores are currently the dominant calcifying organisms in the Mediterranean waters and are expected to play a significant role in global seawater $CO₂$ buffer and in oceanic sink for anthropogenic CO2. The isotopic fractionation in the coccoliths can reflect the biomineralization process and physiological response of calcifying organisms to environmental conditions.

Seawater samples were collected from six depths at stations 1, 3, 5, 7, 9, 11, 13, 16, 18, 20, 22, 24, 26 and 28, for coccolithophore and d^{13} C-DIC analysis. 3 liters of seawater were gently filtered through 0.8 μm pore-size, 25 mm diameter, polycarbonate membranes using a vacuum pump with lower than 20 mm Hg pressure. Filters were oven dried at 60°C and stored in plastic petri dishes. Coccolithophore samples will be analyzed using scanning electron microscope at Xiamen University. d¹³C-DIC will be determined at Woods Hole Oceanographic Institution using a Picarro CRDS (G2131-i) coupled to Picarro Liaison interface and a modified AutoMate autosampler (Subhas et al., 2015).

5.27 Plankton Sampling with Net Deployments

(T. Guy-Haim, M. Gilboa)

Mesoscale eddies form unique habitats characterized by distinct physical and chemical conditions, with high (cyclone) or low (anticyclone) productivity, supporting and distributing a wide variety of plankton, including the larvae and eggs of benthos and fish. The Eastern Mediterranean Sea (EMS) fosters complex and persistent eddying activity. Eddies are of particular importance in the EMS, a "blue desert" characterized by low biomass and productivity levels, accelerated warming due to climate change, and high rates of biological invasions. To test the role of eddies as unique habitats and transport vectors of plankton, we aim at characterizing the micro, meso and macro -plankton within cyclonic and anticyclonic eddies.

Plankton communities were sampled using three consecutive vertical net deployments: (**1**) WP2 net (Hydro-Bios, Ø=57 cm, 50-µm mesh size), (**2**) WP3 net (Hydro-Bios, Ø=113 cm, 1000-µm mesh size), and (**3**) MultiNet Midi (Hydro-Bios, 50X50 cm, 200-µm mesh size, equipped with CTD, fluorometer and oxygen sensor). At each station, the following depth layers were sampled:

The nets were hauled at 0.5 m/s (0-400 m WP3 nets were hauled at 0.2 m/s every alternating day). Filtered water volume was calculated using a mechanical flow-meter (Hydro-Bios). In total, 210 net samples were collected.

| Analysis type | Biomass / SIA / | Metabarcoding | ZooScan/ |
|---------------------------|-------------------------------|----------------------|---------------------|
| | HM | | Planktoscope |
| Sample preparation | Filtration, 55° C 24 | Ethanol 100% | Buffered formalin |
| | h | | (4%) |
| WP ₂ | 50% | 25% | 25% |
| $WP3*$ | 25% | 50% | 25% |
| MultiNet (net $#5$ – net | 25% | 25% | 50% |
| #1 | | | |

Motoda splitter (Hydro-Bios) was used to split each sample as follows:

*SIA=Stable Isotope Analysis, HM=Heavy Metals.

Samples for biomass were filtered on pre-weighted pre-combusted GF/C filters (Whatman) and oven-dried at 55 °C for 24 hours onboard. Following weighing in Guy-Haim Lab, the filters will be halved for C and N stable isotope analysis (Cornell University Stable Isotope Laboratory), and heavy metal analysis (Chemistry dept., IOLR). DNA from the ethanolpreserved mixed samples will be extracted in lab, and used for amplification and sequencing of COI and 18S v9 amplicons (Guy-Haim Lab, IOLR). Additional specimens, manually picked from nets, will be morphologically identified using microscopy, followed by DNA barcoding. Sample (mixed and individual) sequences will be uploaded to NCBI GenBank. Fresh samples from the WP2 net were sieved to obtain the 50-200 µm fraction, and imaged onboard using Planktoscope v2.5 (Fairscope). Following segmentation, all vignettes (e.g., Fig. 2) will be uploaded to EcoTaxa and classified to taxonomic groups using AI (https://ecotaxa.obs-vlfr.fr/) with the corresponding sample metadata. MultiNet $(>200 \,\mu m)$ and WP3 $(>1000 \,\mu m)$ formalinfixed samples will be scanned in Guy-Haim Lab, segmented using ZooProcess and classified to taxonomic groups using EcoTaxa.

Fig. 5.07 Vignette examples obtained by Planktoscope from 50-200 μ m size fraction of 50- μ m mesh WP2 nets hauled to 0-200 m during M197.

5.28 WP3 Net Deployments

(N. Hansen, T. Guy-Haim, M. Gilboa)

Zooplankton nets are one of the oldest oceanographic instruments and are used to study diversity, abundance, biomass, and distribution of zooplankton. During M197, we performed vertical WP3 tows at 0.2 m s^{-1} heaving speed using a 1000 μ m mesh size net from 400 m water depth to the sea surface. The low heaving speed allowed us to sample well-preserved fragile organisms, including gelatinous zooplankton. We equipped the net with a flowmeter to monitor the sampled water volume. From the samples, we picked gelatinous individuals and stored them at -80°C for DNA barcoding and DNA metabarcoding of the stomach contents. At the end, we want to alter the regional DNA barcode library of gelatinous organisms for other approaches, including eDNA. From the stomach content analysis, we will study the trophic ecology of these organisms. Additionally, we picked fish larvae that were stored at 99% EtOH. In total, we had 15 WP3 deployments at 15 station with a total of 100 samples.

5.29 Environmental DNA (eDNA)

(N. Hansen)

We sampled eDNA at 8 stations (M197 2-4, M197 5-5, M197 7-5, M197 9-5, M197 13-5, M197 18-5, M197 20-4, M197 26-4) by filtering 2 liters of water in triplicates (3 filters) each at 6 ecological depths We took negative controls (MilliQ water) at each station. We sampled a total of 141 eDNA Sterivex[™] filters from in situ pump CTD water and 8 eDNA Sterivex[™] filters from MilliQ water. We will apply DNA metabarcoding protocols to identify species that avoid towed instruments and that we do not see in pelagic video transect nor catch with zooplankton nets. We will extract the DNA from the filters via standardized extraction kits, then perform polymerase chain reactions (PCRs) to amplify the DNA region of interest. The amplified DNA will be sequenced via Next Generation Illumina Sequencing. Furthermore, we also sampled sediment from the upper 1 cm of cores taken with the TV Multicorer at 16 stations. With these samples we will also apply DNA metabarcoding to identify pelagic species that may play a role in benthic-pelagic coupling.

5.30 In Situ Camera – Small Pelagic in situ Observation System

(N. Hansen)

During M197, we were interested in the vertical distribution of gelatinous zooplankton in the epipelagic (0 - 200 m) and mesopelagic (200 - 1000 m). The abundance, diversity and distribution of organisms in the water column can vary with sea surface productivity, stratification (i.e., the ecological zones that are shaped by the physical and biogeochemical environment), and temporal patterns, including seasonal and diel dynamics. For example, the distribution of certain taxa varies between day and night as a result of diel vertical migration (organisms are hiding in the mesopelagic during day and migrate to the sea surface to feed during night). Due to their fragile nature, most gelatinous organisms are destroyed by traditional zooplankton sampling methods, including nets. Therefore, in situ optic systems are used congruently to zooplankton nets to study gelatinous zooplankton. We deployed the small towed pelagic in situ observation system Baby-PELAGIOS (Fig. 5.08) for horizontal transects

at 9 distinct depths between 25 m and 900 m. The Baby-PELAGIOS was equipped with a forward-looking camera (SubC 1Cam) and 4 LEDs illuminated the water in front of the camera. The camera recorded video with a resolution of 1440x1080 at 25 fps. We monitored the water depth with a HYDRO-BIOS pressure sensor that was mounted on the frame. We performed the horizontal tows at 1 knot (0.52 m s^{-1}) over water of 12 minutes. We selected the transect depths according to the following ecological zones that were identified using provided CTD and Parasound (18kHz) data; mixed layer, deep-chlorophyll maximum (DCM), thermo- and halocline, oxygen minimum zones and scatter layers. In total, we had 24 PELAGIOS deployments (12 day and 12 night) at 12 stations and collected 60 hours of video.

The imagery data will be transformed into iFDOs (image FAIR Digital Objects), containing the video files and all relevant metadata, and stored on the ELEMENTS Server at GEOMAR. At GEOMAR, we will identify the observed taxa (i.e., annotation) to infer the vertical distribution and abundance of gelatinous zooplankton in the eastern Mediterranean Sea. Furthermore, we will test and use image process tools, including segmentation and object detection, to support annotation. At the end, we aim to correlate the vertical distribution, diversity and abundance of gelatinous taxa with e.g., the presence of mesoscale features such as eddies in the eastern Mediterranean Sea.

Fig. 5.08 The small pelagic in situ observation system (Baby-PELAGIOS) equipped with 4 LEDs, the forward-looking SubC 1Cam camera, HYDRO-BIOS depth sensor, sea anchor, and weight.

5.31 Aerosols and Bioaerosols Sampling

(B. Herut, B. Ankri)

Aiming to investigate the diversity, abundance and viability of airborne microbes and their associated aerosol chemical composition and interactions with seawater, the following measurements were performed (See also Section 12.16 for schematic):

Bioaerosols low volume sampling - Low volume (8 L min−1) air collection was performed using BioSpot-VIVAS (Aerosol Devices Inc., Fort Collins, CO) that condense aerosols (and its corresponding airborne cells) into a petri dish (Pan et al., 2016; 2018) containing sterile 0.2 um filtered surface seawater (35 mm, 4 ml). The four major sections of the device are the conditioner, initiator, moderator, and collector, which were operated at 4 °C, 40 °C, 28 °C, and 25 °C, respectively. The collected cells were stained and preserved (see below details), thus enabling us to follow the aerosol-borne cells' abundance and viability. The instrument was positioned in the meteorological laboratory with its intake tube on the monkey island of the vessel (no smoking area). Aerosols were collected ~twice per day, 10 h each run continuously during the cruise. Subsamples were collected for prokaryotic bacterial abundance (LIVE/DEAD), nutrients and DNA (if possible). For the prokaryotic bacterial abundance (LIVE/DEAD) water samples were stained immediately upon collection with a mixture of two nucleic acid stains; green-fluorescent SYTO®9 dye and red-fluorescent propidium iodide, and will be analyzed by an Attune® Acoustic Focusing Flow Cytometer (Applied Biosystems) equipped with a syringe based fluidic system and 488 and 405 nm at a flow rate of $25 \mu l$ min-¹. The collected samples were kept at 4 °C. For the nucleic acids (DNA level) available volumes of water were filtered through 0.2 µm sterile PALL filter. The collected filters were kept frozen -20 \degree C until analyses. A total of \sim 54 samples were sampled during the cruise.

Aerosols High volume sampling - Two high volume aerosol collectors were operated, one for PM10 aerosols fraction and the 2nd for total suspended particles (TSP) in air. Both samplers were located at the monkey island of the vessel (no smoking area). For PM10 the flow rate was set at 67.8 m³ h⁻¹ and aerosol collection was performed on Whatman 41 filter (acid-cleaned, 203 mm x 254 mm) for DNA, leachable nutrients and other related trace elements. Filters were usually changed every 36-48 h and stored frozen (−20 °C). For TSP the flow rate was set at 70 $m³ h⁻¹$ and aerosol collection was performed on Quartz (QMA) filter (203 mm x 254 mm, preheated to 450 °C for 3 h) for DNA, chemistry and potentially biotoxins. Filters were changed every 48 hrs and stored frozen (−20 °C). The sampling started on 6 January 2024 and finished on February $4th$, 2024. A total of 16 filters per collector were sampled during the cruise.

Sea-spray sampling - Sea-spray aerosols were collected using a bubble-generating system (Rastelli et al., 2017). Surface $(\sim 5 \text{ m})$ seawater from the RV METEOR Membrane pump were flowing throughout the cruise track into an airtight bubble bursting tank (200 L). A bubblegenerating system was applied at seawater flow rate of 20 L min-1 to produce bubbles, and, in turn, bursting at the water surface forming sea spray aerosol (estimated production of $\sim 2.5 \times 10^7$ particles m^{-2} s⁻¹). Such a system has been shown to produce bubble size spectra representative of the breaking waves at open sea and to generate sea-spray particle size distributions representative of the natural ones (Hultin et al., 2010). The collected aerosol particles are concentrated into a petri plate containing 15 ml sterile seawater $(< 0.2 \mu m$) mounted on a stage Berner impactor operating at a flow rate of 80 L min⁻¹. Daily 6 h collection was performed. Subsamples were collected for prokaryotic bacterial abundance (LIVE/DEAD), nutrients and DNA (if possible). The collected material was fixed and stained for LIVE/DEAD analyses and total bacterial abundance reads using flow cytometry. A total of \sim 30 samples were sampled during the cruise.

Microcosm experiments $- A$ set of 5 microcosm experiments were performed following different treatments. The experiments included the addition of dust/aerosols collected during a Saharan dust event on the 8-9.1.2024; additions of nutrients; additions of isolated airborne

autotrophs and copepods. The microcosm experiments were performed using surface seawater (5 m deep) collected by the tow-fish pump at stations 2, 3, 9,16 and 24. Incubations were performed onboard in 250 ml pre-cleaned (acid washed) transparent bottles located in a seawater flow tank for 24-50 hours. Subsamples were collected for prokaryotic bacterial abundance (LIVE/DEAD), alkaline phosphatase activity (APA), phytoplankton photophotophysiology, nutrients and DNA (if possible). Subsamples of 0.5 ml were fixed and stained for LIVE/DEAD analyses and total bacterial abundance reads using flow cytometry. Subsamples of 15 ml were run onboard (I Bermn-Frank) by Chelsea Technology Ltd LabSTAF instrument for single-turnover active fluorometry (STAF) used to assess phytoplankton photophotophysiology and estimate photosynthetic rates. Subsamples of 10 ml were run onboard for APA.

5.32 Underway Hydroaccustics

(H. Wilckens and O. Bialik)

Multi-Beam Echo Sounder: During M197, the seafloor morphology was mapped with a Multi-Beam Echo Sounder (MBES), the Kongsberg EM122. The MBES Data acquisition was carried out along the cruse track inside the study area. The MBES is permanently installed on the ship's hull and operated at a frequency of 12 kHz. The swath opening angle across track was set to 130° and the opening angle of each of the 400 beams is 1 x 2 degrees. The Beam Spacing was set to HD Equidistance. For accurate determination of the depth the sound velocity profile was calculated from the CTD data at the stations 1, 21 and 22. Both seafloor and water column data have been acquired and stored. The MBES was very reliable throughout the cruise. On board, the data has not been processed. The data will later be processed and analysed at the University of Haifa. RV METEOR is equipped with a second MBES the Kongsberg EM 710 for shallow to mid-water. The MBES was tested for a short time for imaging the water column but was than permanently turned off due to cross talk between the instruments.

Sediment Echo Sounder: The parametric Sediment Echo Sounder (SES) Parasound DS-3 from ATLAS HYDROGRAPHIC GmbH can image the sub-seafloor structures up to 200 m depth in high resolution. The SES is permanently mounted on the hull of the RV METEOR. The SES makes use of the parametric effect to produce a secondary low frequency based on two primary high frequencies. During M197 the secondary low frequency was set to 4 kHz. The primary high frequency and secondary low frequency was stored as *.asd files and exported to PS3 to SEGY format and visualised with SeiSe of initial quality control. The primary high frequency was also used for imaging the upper 1000 m of water column to show the depth interval of plankton and the migration of zooplankton. This information was used for planning the target area of the PALAGIOS. The system worked mainly reliably and continuously in the designated study areas during the cruise. Once the system was unable to find the seafloor do to steep topography changes leading to a small data gap. Detailed processing and analysis will be conducted after the cruise.

Fig. 5.09 A) Example the unprocessed MBES 122 showing the Napoli Mud Volcano. B) Photo of brine pool with camera that was mounted on top of the Multicorer.

Vessel mounted Acoustic Doppler Current Profiler: During M197 two vessel mounted Acoustic Doppler Current Profiler (ADCP) from Teledyne RD Instruments were operated continuously in the Cypriot EEZ and in ide the designated study area in the Greece EEZ. The ADCP makes use of the Doppler shift to measure current speed and direction in the upper water column. Ocean Surveyor 38 kHz was installed in the on the ship's hull. Due to a malfunction the 38 kHz ADCP only started to work during Station 1. The 38 kHz was setup to high resolution narrowband and the broadband single-ping profile mode. The Bin size was 16 m with 80 bins in total. The blank distance was 8 m. This led to recording of the upper \sim 1200 m from the water column. Ocean Surveyor 75 kHz was installed in the moonpool instead of the hydrographic shaft. The switch in position probably increased the data quality during recording on station but lowed the resolution during sailing. From the 19.01.2024 to 21.01.2024 the 75 kHz was swished of because the POSIDONIA system was installed in the moonpool. The 75 kHz was setup to low-resolution, long-range profile and the narrowband single-ping profile mode. The Bin size was 8 m with 100 bins in total. The blank distance was 4 m. This led to a recording of the upper ~800 m from the water column. The ADCP data was processed during the cruise with the Cascade V7.2 software and displayed with MATLAB (Fig. 5.10). The data will be analyzed in detail after the cruse to better understand the currents during the cruise and the migration of plankton.

Fig. 5.10 Example of hydroacoustic data collected in the water column during Station 9 for A) Sediment echosounder primary high frequency of 18 kHz, B) 75 kHz ADCP showing the eco intensity, C) 38 kHz ADCP showing the northward, D) eastward and E) vertical velocity.

5.33 Mesoscale Eddy Identification

(M. Gilboa)

Throughout M197 expedition we characterized semi ephemeral and semi constant oceanic features such as mesoscales eddies, with time scales of days to months and mean spatial scales of ±35 km in the Eastern Mediterranean Sea. Such features can sustain oceanic conditions that promote distinct planktonic communities. In the Mediterranean, anticyclonic mesoscale eddies have a clockwise circulation where warn and nutrient poor sea surface waters are down welled. In comparison, cyclonic eddies can upwell nutrient rich colder waters from deeper layers, enhance primary productivity and support higher planktonic biomass. To better understand how plankton dynamics and community structures are affected by mesoscale eddies, we aimed to sample as many features as possible. This was achieved by near real time on-board analysis. The eddy detection based on daily satellite altimetry (Copernicus, SEALEVEL_EUR_PHY_L4_NRT_OBSERVATIONS) and consisted of derived geostrophic velocities from sea surface height (SSH) to calculate and detect the sea level anomalies compared to absolute dynamic topography. These anomalies were then plotted together with streamlines around minimal and maximal values and then detected as eddies. On a regular basis the plots were used to aim towards areas of interest. The corroboration of eddy existence, structure and intensity will be achieved using the physical water characteristics that were measured in situ within and outside of these features. An example figure is shown in Section 12.17.

5.34 Multicore Deployments

(W. Bett, O.M. Bialik, H. Wilckens, Z. Steiner)

Sediments and porewaters: The sediment is an integrated archive of material which were transported to and form in the water column. An exception to this was encountered in stations 14 and 16 on the Napoli Mud Volcano where material is advected from the subsurface onto the seafloor. This material was modified by chemical reactions and microbial activity through its accumulation and burial. These transports and reactions make the sediment both a sink and a source for different elements. In the open oligotrophic waters of the Eastern Mediterranean, the sediment accumulates biogenic material from the water column and benthic community as well as lithogenic material derived from the surrounding continents.

Porewaters are the aqueous medium in the sediment. Diffusion and advection between the porewaters and the bottom waters is an important vector of transport of dissolved material to the deep sea. Porewaters are also a convenient medium for the study of water-solid interactions and the effects of microbial activity. These diagenetic processes are most active within the top of seafloor sediments during accumulation and are driven by organic matter respiration and limited replenishment of oxygen, a combination that often leads to the formation of anoxic conditions in the porewaters.

During cruise M197 short sediment cores with overlaying water were collected using the GEOMAR multi-corer (MUC) with video-telemetry (Fig. 5.11a). The MUC was loaded with 11 liners (60 cm long, 10 cm diameter), three of which were pre-drilled for porewater extraction using Rhizon samplers. Sediment cores were successfully retrieved at 20 stations during the cruise. The last 100 m of the descent were recorded using the video-telemetry to visualize the seafloor surface and ensure safety of decent. The video-telemetry showed the lower part of the liners (Fig. 5.11b) in the MUC and was used to observed if the MUC closed in the water column during high waves. The objectives of the sediment work done during the cruise are to quantify nutrient, trace and major element fluxes between the sediment and bottom-water, explore the benthic environment of the Eastern Mediterranean, and reconstruct changes in the regional ecosystems and pollution levels in the recent past with a focus on the last centuries. A summary of collected sediment samples is provided in Section 12.18.

Fig. 5.11 (A) MUC on deck with sediments from station 9. There is a layer rich in pteropod shells at the surface, and a sapropel at the bottom of the core. (B) Video Telemetry of the seafloor from station 26

Porewaters: The best predrilled core liner, with a visually undisturbed surface and filled to the top with clear overlying water was selected and moved to a cold room shortly after retrieval. The temperature in the cold lab was set to match the Eastern Mediterranean bottom water temperature of 14°C, and the cores were initially left to re-equilibrate to bottom water temperatures for 1-2 hours. The Rhizon samplers were pre-cleaned by soaking them in 3 nM HCl for 24 hours and rinsed and soaked in milli-Q water for a few days before use. The overlying water was sampled using HCl cleaned syringes and filtered using 0.2 μm PES filters. Before collection of overlying water samples, the syringe, filter, and vials were rinsed with the sample three times. 10 mL was each collected for nutrients (analyzed on-board), trace and major elements, and 1 mL for Ammonia. The Ammonia samples were frozen in −20 °C. The rest of the overlying water was then drained. In the first 3 stations, additional samples for alkalinity were also collected. But on-board nutrient results of the overlying water showed values similar to shallow-water depths, indicating contamination of the water during ascent of the MUC. Subsequent alkalinity collection was therefore not performed. At the brine pool stations, overlying water from 4 additional cores was collected for nutrients, trace and majors, alkalinity, chlorinity, and mercury.

The Rhizon samplers were inserted into the drained core and samples collected from top to bottom, usually at a 2 cm resolution. In longer cores, the first 10 Rhizons were inserted every 2 cm and afterwards the resolution decreased to every 3 cm. The first 1 mL collected by each of the Rhizon samplers was discarded, and the next Rhizon inserted only after this step was complete. At total of 5 mL was collected using each Rhizon sampler, 1 mL was set for ammonium and frozen until analyses (−20 °C), the remaining water was stored in HCl cleaned 5 mL polypropylene (PP) tubes and acidified with 50 μL of 4 M Supra HCl at stations 1-12, and from station 13 onwards with 25 μL of concentrated Supra HCl. The tubes contained between 2 and 5 mL of solution. Porewater nutrient samples were diluted 1:6 with nutrient depleted surface seawater sampled from the tow fish. Both the porewater and overlying water were analysed on board for concentrations of soluble reactive phosphorous, nitrite + nitrate (TON), nitrite, and silicic acid. For major element analysis (Ca, Mg, S, K, Sr, Li), samples were diluted at a 1:100 ratio with 0.12 M Supra HCl. The major element concentrations will be analyzed at GEOMAR for their element to sodium ratios using a Varian 720 ICP-OES (Inductively Coupled Plasma Optical Emission Spectroscopy). Trace element samples were diluted 1:25 with 1 M distilled $HNO₃$, and will be analysed at GEOMAR using an Element-XR HR-ICP-MS (High Resolution Inductively Coupled Plasma Mass Spectrometry).

Core incubations: Sub-cores 3 and 4 were designated for incubation experiments to measure fluxes of inorganic nutrients and major elements at the sediment-water interface. The experiments were carried out in the cold room (14°C) in dark conditions. Cores from the first 2 stations were left to equilibrate for 24 hours before the experiment began. In later cores, the experiment started between 8-12 hours after retrieval. An inlet pipe was suspended a few centimetres above the sediments and a shorter outlet pipe submerged below the top level of the water. A peristaltic pump was then utilized to maintain a continuous flow system in both cores. Samples were collected at the start of the experiment (at time T0) to have a baseline of the concentrations. Subsequent samples were collected through a costume made T port at intervals of every 6 hours for the first 4 samples and later increased to every 8 hours for the last three samples. 5 mL was retrieved for nutrients (analyzed on-board), 5-10 mL for trace and major elements. In each sampling between 3-4 mL were discarded during the syringe and filter rinse before sample collections.

Four additional incubation experiments were carried out with resuspended sediment. Sediment from stations 19, 22, and 27 taken from core depths of 0-1 and 9-10 centimeters down cores was resuspended in 0.2μm filtered deep water. Samples from three of the experiments were sampled and filtered every few hours through the experiment for nutrients and trace metals through the 24h run of the experiment. Nutrients from the first two experiments (except ammonia) were measured onboard, and nutrient samples from the third experiment were frozen for analysis in GEOMAR post-cruise. The fourth experiment was a 10h batch experiment with addition of isotopically labeled nitrogen species (N2, NO3, and NH4), which will be analyzed for nitrogen utilization post-cruise.

Solid sediment: Subcore 5, selected as the most representative core, longest and with undisturbed surface was designated for physical properties (porosity and density) and geochemical analyses of clays and bulk mineralogy, trace and major elements, organic carbon and dating techniques. These samples were sliced at a resolution of 0.5 cm for the top 5 cm, 1 cm from 5-15 cm and 2 cm from 15 cm to the total depth of the core. The outer part of the core was collected and stored in plastic zip lock bags, while the inner part was in pre-weighed acrylic bottles. The later will be analyzed post-cruise for porosity and density. In the last three station the outer part of the liner was discarded and the rest was collected into 80 ml plastic containers.

Sub-core 6 was designated for micropaleontological analysis and was sliced at 1 cm resolution for the entire depth. The top 6 cm were sampled for living foraminifera. The samples of living foraminifera were collected in ~80 ml plastic containers with 2/3 of ethanol added to 1/3 of the sediment sample to preserve it. Below this depth, samples were collected at 1cm resolution and stored in plastic bags. All treatment and analysis of these samples will occur post-cruise.

Additional micropaleontological samples targeting pteropod were collected if additional sub-cores were available. This sub-core was taken at various stations and slicing was carried out based on observation of pteropods during slicing of other sub-cores. Pteropods are diverse planktonic gastropods that occupy unique habitats in the ecosystem and represent an excellent proxy for ocean acidification due to their aragonite shells. The samples collected were washed on-board over a sieve of mesh size 250 μM and transferred to an oven set at 40° to dry. Subsequent analysis of these sample will be carried out post-cruise with emphasis on their mineralogy and geochemistry.

The top 5 cm of subcore 7 was sliced with a metal tool and stored in a glass jar for analyses of microplastics (see 'Microplastics' section).

The top surface of subcore 8 was sampled with sterile tools for eDNA analysis. In some stations samples for Metagenomics were collected from this core at various depths and later stored in the −80°C freezer.

For visualization of the cores using CT, a 50 mm diameter plastic tube (cut to sediment length + 1 cm) was centrally inserted into the longest undisturbed core, which was then capped. In some stations, samples of Mercury were collected from this core at various depths into small petri dishes and later stored in the −20°C freezer. Mercury samples were collected from a away from both the subcore and the core liner as possible.

All the sediment samples for micropaleontology and geochemistry were stored in a cold room (4°C) after slicing.

5.35 Underwater Vision Profiler

(T. Guy-Haim, M. Gilboa, C. Eckmann)

UVP6-HF installed on the rosette was operated in "CTD" mode. The UVP casts were obtained from each station and uploaded to EcoPart under uvp6_sn000159hf_202301_m197 project. Profiles will be analyzed based on size fraction and corresponding environmental parameters (e.g., Section 12.19). UVP vignettes (segmented images) are uploaded to EcoTaxa and classified to taxonomic groups based on local taxonomic expertise and corresponding stations net samples.

6 Ship's Meteorological Station

(P. Suter)

On the morning of 6 January, Meteor left the port of Limassol heading south in calm seas and sunny, high-pressure weather with weak winds.

A high-pressure zone over the southeastern Mediterranean began to weaken and the research area south of Cyprus was situated on the downstream area of a large low-pressure complex over Italy by 8 January. With slightly increasing winds from the south to southwest, the weather remained calm with temperatures of up to 20°C. However, advected Sahara dust caused slight turbidity in the air. The low-pressure zone spread eastwards over Italy and formed a secondary low in the Aegean Sea by 9 January, which then moved eastwards along the southern Turkish coast. The westerly current reached 6 Bft from midday on 9 January. In conjunction with a cold front in the early morning of 10 January, there were some heavy showers with lightning in the vicinity of the ship as well as gale-force gusts. With the inflow of drier air in the middle and higher layers, the shower activity decreased again during the day. At the same time, the waves from west rose to 2.5 to 3 m.

Subsequently, a stationary low-pressure zone persisted over Cyprus and the Taurus. As a result, it remained windy on 11 and 12 January with winds from west to northwest between 4 and 7 Bft and changeable clouds as well as intermittent showers. The sea reached wave heights of 2.5 to 3.5 m. By 13 January, another low-pressure system followed from Crete, which moved just north of the Meteor towards Cyprus. The passage of the low brought strongly fluctuating winds in the warm air sector in conjunction with repeated showers, with average winds of 6 to 7 Bft at times and gale-force gusts of around 40 kt. At the same time, the lowest air pressure of the expedition was measured with 1006.3 hPa. In the evening, a cold front brought in drier air. The weather and the sea calmed down.

By the evening of 14 January, the research work south of Cyprus was completed within a filling low-pressure zone and Meteor moved westwards into the southern part of the Taurus sea area. Towards the east, a high-pressure bridge formed on 15 January, connecting two highs over eastern Turkey and Egypt. The northwesterly to westerly current was still blowing with moderate to fresh forces. Repeated sunny spells alternated with widespread cloud cover and isolated showers. A new low had formed over northern Italy in the meantime and so the current turned back to southwest by the evening of 15 January. By 16 January, the low-pressure zone had spread eastwards over Italy and formed a secondary low in the Aegean. Between the highpressure bridge in the east and the secondary low, Meteor was situated within a stormy southwesterly current at times. The wind reached up to 36 knots in the morning, with gusts up to 48 knots. The sea temporarily built up to 3.5 m.

From 17 January, the secondary low moved eastwards along the southern Turkish coast. As a result, Meteor remained between higher pressure to the east and a new low-pressure zone over the western Mediterranean. Thus, calm and, apart from denser cloud cover on 20 January, mostly sunny conditions accompanied the research work southeast and south of Crete until 20 January.

On the night of 21 January, a new low-pressure system moved eastwards from Sicily just north of Meteor over Crete. In conjunction with a high-pressure zone over the Balkans and a marked air pressure gradient, gale-force winds from the north blew in over the Aegean and as far as west of Crete. Overall, the ship was often sheltered from the stormy winds and high seas southwest of Crete until 24 January. However, due to the weather forecast, the station for 22 January was changed in order to avoid the high waves in the southwest of Crete. At the same time, Meteor also benefited from mostly sunny and dry conditions in the lee of the island. The observed wave heights were between 2 and 2.5 m.

On 25 January, Meteor moved slightly westwards to the southwest of Crete and thus left the area protected by the island. A fresh to strong northwesterly wind was still blowing. However, together with the significantly lower waves, the conditions were not comparable to the last few days at this position. On 26 and 27 January, Meteor moved in the southeastern part of the Ionian Sea. Accompanied by plenty of sunshine, a mostly weak northwesterly- and a northeasterly current flowing out of the Aegean repeatedly alternated here.

On 28 January, a strong high-pressure system moved from Central to Eastern Europe and a stationary low-pressure zone remained near Cyprus. Between the pressure formations, a large pressure gradient built up in the Aegean and Ionian Sea. In the area of a convergence zone, the initial northwesterly wind was abruptly replaced by an east-northeasterly wind coming from the southwest Aegean. This quickly increased to 8 Bft with a maximum at 36 knots and only decreased slightly to 7 Bft in the afternoon. The sea increased rapidly in the afternoon hours to around 3 m, with individual waves over 4 m. Showers formed with the converging currents and due to a marked wind shear in the lower air layers and the inflowing cooler air, two welldeveloped waterspouts were also observed. By 30 January, the high had shifted from Ukraine to Romania and remained stationary there. At times there was a pressure difference of 30 hPa between the high and the intensifying low near Cyprus, with Meteor recording the highest air pressure of the trip at 1030 hPa on 29 January. Accordingly, the stormy northeasterly winds persisted in the Aegean and as far as south of Crete. Meteor was already slightly to the west of the strongest winds, with mostly around 6 Bft from northeast to east and wave heights of 2 to 2.5 m prevailing until the morning of 30 January. During the course of 30 January, Meteor was temporarily in the lee southwest of Peloponnese with waves of around 1.5 m before a strong northerly current broke through again in the evening. With the advected cooler air from Eastern Europe, many showers developed in the unstable air mass and small hail also occurred with a cloud pattern that changed very frequently. The launched radio sounding suggested a snowfall line of 600 to 700 m and the lowest temperature of the expedition were recorded here with 8.5°C during a shower.

From 31 January, the high-pressure zone over the Balkans and the low-pressure zone near Cyprus weakened and the differences in air pressure over the eastern Mediterranean decreased. The air mass also stabilised and warmed up again. In the Ionian Sea, the influencing pressure systems were only weak for the last few days of the expedition. The winds came most of the time from northwest to northeast with 2 to rarely 5 Bft and the waves were also only slightly pronounced. In the morning hours of 6 February, Meteor arrived in Catania in sunshine and light winds.

7 Station List M197

7.1 Overall Station List

EM122= Deep-sea Multibeam Echosoundser; ADCP= Acoustic Doppler Current Profiler (75 or 38 kHz); PS= Parasound; UAS= underway air sampling; CTD= Meteor CTD; CTD-UC= CTD Ultra Clean; ISC= In situ Camera (PELAGIOS); TV MUC= Video Multicorer; WP2= WP2 plankton net; WP3= WP3 plankton net; MSN= Multinet; Pump= tow-fish; ISP= in situ pumps.

8 Data and Sample Storage and Availability

A cruise summary report (CSR) has been compiled and submitted to DOD (Deutsches Ozeanographisches Datenzentrum), BSH, Hamburg, immediately after the cruise. Part of the cruise was performed in waters under jurisdiction of the Republic of Cyprus and part under jurisdiction of Greece.

All hydrographic data acquired during the cruise will be transferred to BSH and made available to the PANGAEA database. All nutrient and trace element to be acquired will also be fed into these data bases and will be made publicly available within 3 years after cruise end $(1st$ quarter of 2027). All water samples are stored at the respective laboratories, where the measurements will be carried out. The Kiel Data Management Team (KDMT) provides an information and data archival system where metadata of the onboard DSHIP-System are collected and are made publicly available. This Ocean Science Information System (OSIS-Kiel) is accessible for all project participants and can be used to share and edit field information (https://portal.geomar.de/metadata/). Metadata will be made available in OSIS (https://portal.geomar.de/osis) following completion of the cruise, whilst the data will be made available in OSIS according to Table 8.1.

Table 8.1 lists the target databases, tentative availability times and responsible scientists.

Hydrography and hydroaccustics - data are held by DAM and will be publicly available within six months after the cruise (responsible: Dr. T. Browning).

Dissolved and particulate trace metals - samples and data are held at GEOMAR, Kiel (responsible: Prof. E. Achterberg/Dr. T. Browning).

Phytoplankton/productivity – samples and data are held at University of Haifa (responsible Prof. Ilana Berman-Frank), and GEOMAR, Kiel (responsible Dr. T. Browning).

Nitrogen cycling and fixation - samples and data are held at University of Haifa (responsible Prof. Ilana Berman-Frank).

Zooplankton and eDNA - samples and data are held at GEOMAR and University of Haifa (responsible Dr H.-J. Hoving and Dr. T Guy-Haim).

Nutrients, DOC, carbon cycle - samples and data are held at GEOMAR, Kiel (responsible Prof. E. Achterberg/Dr. T. Browning).

Microplastics - samples and data are held at CMMI, Republic of Cyprus (responsible Dr. L. Hadjioannou).

Microbial DNA/RNA - samples and data are held at MBL, USA (responsible Prof. A. Worden).

Sediments - samples and data are held at GEOMAR and University of Haifa (responsible Prof. Y. Makovsky).

| Type | Database | Available | Contact |
|-------------------------------|-----------------|-------------------|--|
| Hydrography | PANGAEA | July 2024 | tbrowning@geomar.de |
| Nutrients | PANGAEA | March 2027 | eachterberg@geomar.de |
| Trace metals | PANGAEA | March 2027 | eachterberg@geomar.de |
| Nitrogen cycling and fixation | PANGAEA | March 2027 | iberman2@univ.haifa.ac.il |
| Phytoplankton/productivity | PANGAEA | March 2027 | tbrowning@geomar.de iberman2@univ.haifa.ac.il |
| Carbonate chemistry | PANGAEA | March 2027 | eachterberg@geomar.de |
| Zooplankton | PANGAEA | March 2027 | hhoving@geomar.de tamar.guy-haim@ocean.org.il |
| Sediments | PANGAEA | March 2027 | yizhaq@univ.haifa.ac.il |
| Microplastics | PANGAEA | March 2027 | louis.hadjioannou@cmmi.blue |
| Microbial DNA/RNA | PANGAEA | March 2027 | azworden@mbl.edu |

Table 8.1 Overview of data availability

9 Acknowledgements

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11 Abbreviations

Appendices

12.1 Total Salinity Samples Taken from SS CTD, Including Station Numbers.

12.2 Total Salinity Samples Taken from TM CTD, Including Station Numbers.

12.3 Underway Samples for Collected Salinity Analysis.

12.4 Total Oxygen Samples Taken with SS CTD

12.5 Total Oxygen Samples Taken with TM CTD

12.6 Total Oxygen Samples Taken from Cores Incubation

12.7 Total Oxygen Samples Taken for EXO Oxygen Sensors Calibration

12.8 Samples for Dissolved Thorium and Uranium

12.9 Samples Taken from the In-situ Pumps.

(particulate thorium (pTh), particulate inorganic carbon (PIC), particulate organic carbon (POC), particulate organic phosphorous (POP), biogenic silicate (BSi), genomics, aminoacids (AA), aminosugars (AAS))

12.10 Station Information for DOM-POM Samples.

12.11 Station Information of APA and POP Samples.

12.12 Information of DOP Incubation Experiment Setup.

12.13 Summary of Measurements/Protocols Taken for the LabStaf Instrument

12.14 Concentration of Added 15N Tracer and Assumed Ambient N Concentration.

In this table the actual measured concentration is shown together with the final percentage of ¹⁵N tracer from the total amount of each N species in the original sample. Note that in all cases except Stations 15, 24 nitrate, which marked in red, the tracer was less than 20% of the ambient.

12.15 N and P Compounds and the Actual Concentrations Added in Each Bottle.

12.16 Aerosol Sampling

12.17 Mesoscale Eddy Detection

Fig.1. Example of eddies detected during the period of the 7-13th of January, 2024

12.18 Multicore Station Report

Multicore station

12.19 Underwater Vison Profiler Profiles From Three Casts in Station 15