MORRIS KAHN MARINE RESEARCH STATION

אוניברסיטת חיפה
University of Haifa
جامعة حيفا

LEON H. CHARNEY SCHOOL OF MARINE SCIENCES
בית הספר למדעי הים על שם ליאון צ'רני

Report for activity years 2015-2021 |Seasons 1-10

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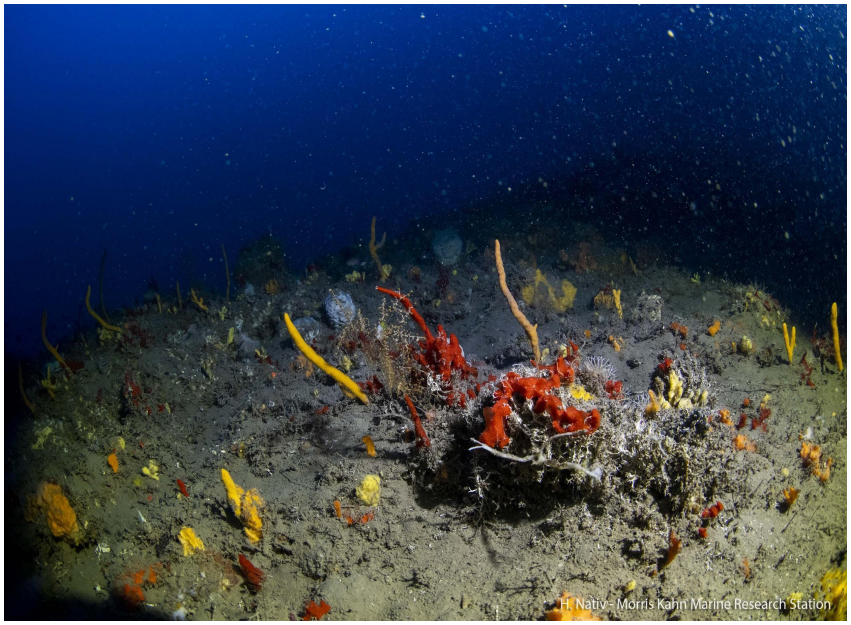
Short-beaked common dolphins (*Delphinus delphis*): a mother and pup off the coast of Ashkelon

Credit: Aviad Scheinin

2021 Report Executive Summary

This report is a summation of 6 years of work since MKMRS was first established in 2015, and consists of the results from its long-term research of the Eastern Mediterranean Sea (EMS) ecosystem (2015-2021).

The Eastern Mediterranean Sea in general, and the Israeli coastal shelf in particular, is a unique ecosystem. The EMS is a semi-enclosed sea with rather large inputs of externally supplied nutrients (nitrate and phosphate), yet the EMS has among the lowest levels of nutrients of any ocean in the world. The EMS coastal ecosystem, also unusually oligotrophic compared to other shelf systems, has been subject to major environmental changes, chiefly the completion of the Aswan Dam in 1965 which stopped the annual Nile flood and reduced incoming nutrient fluxes, and the building and widening of the Suez Canal; this led to it becoming an autostrada for alien species. These pressures are combined with fisheries, and climate change impacts which are causing the surface waters to become warmer, saltier and more acidified compared to other marginal seas.



Sponge garden located at 85 m depth near Nahariya

In view of these processes, the Morris Kahn Marine Research Station started collecting data on various oceanographic and biodiversity parameters, with the aim to widen the available scientific data and make it available for advancing science-based management of the Eastern Mediterranean Sea. The following is a summary of the data presented in this report, organized according to the various laboratories within MKMRS: oceanographic parameters, biogeochemistry, bacteria and phytoplankton in the seawater, marine microbiome, zooplanktonic biodiversity, rocky reef biodiversity, apex predators, and marine pathology.

Marine biogeochemistry

The biogeochemistry of the Israeli marine shelf was described in 1986, and has not been studied since (Berman et al., 1986). To understand what drives this ecosystem, we selected five stations for our long-term monitoring: across the shelf from 100 m to 10 m depth, above a sandy seafloor, away from ports and coastal infrastructure, away from coastal stream estuaries, and across from Kibbutz Ma'agan Michael. We began our study in December 2017, analysing inorganic nutrients in seawater (ammonia, nitrate, phosphate and silicate), chlorophyll, salinity, and temperature along a depth gradient. We have continued to monitor these parameters four times a year. Our preliminary results indicate that the coastal shelf behaves similarly to the offshore environment. It has similarly low nutrient concentrations, and is characterised by low chlorophyll and nutrient dynamics. The main source of nutrients comes from upwelling onto the shelf, with less inputs from: (i) streams, especially during storms, (ii) submarine groundwater, (iii) direct waste discharge, and (iv) the atmosphere. On some occasions, high nutrient concentrations were observed originating from the coast and reaching the shallow waters (observed at 10 and 25 m depth sites).

In addition, we have been conducting seasonal cruises and experimental studies in the upper photic zone in the offshore environment (pelagic stations). We have observed systematic, seasonal changes in inorganic nutrients. In winter, the moderate levels of nitrate are caused by deep mixing of nutrient-rich waters decreasing through the summer, while the surface waters are always depleted in phosphate. Our results have confirmed that these waters are behaving more like mid-ocean gyres. In addition, our results mean that these waters represent a natural laboratory for those large areas of the world's oceans, which are both oligotrophic and phosphate-depleted.

Sandy bottom at 5 m depth near Hadera

Sediment microbial communities

The sediment microbiota plays a significant ecological and biogeochemical role in marine

ecosystems, and our knowledge is still very limited, especially along the Israeli continental shelf. We established an initial database from an undisturbed area (Ma'agan Michael). Microbial communities (bacteria, archaea and eukaryote) are characterized twice a year (winter and summer) along a depth gradient from10-100m. Three years of observations show that the sediment microbiota is stable in composition. Bottom depth was mainly associated with a variation in microbiota composition, followed by core depth and season. We identified several bacterial biomarkers characteristic of each bottom depth and season.

Our initial monitoring efforts yielded a steady baseline. From this, we selected several additional sites outside the Ma'agan Michael transect to test our model. The two-factor model tested the control "undisturbed sites" against the test "disturbed sites", of which the latter are exposed to intensive anthropogenic activity. Only samples from the disturbed sites were found to be significantly altered in terms of sediment microbiota. These preliminary results demonstrate that: 1) sediment microbiota present a complex and dynamic system, and our work yielded robust results, making it an ideal model system for studying intra- and inter-kingdom interactions; 2) Sediment microbiota may serve as a highly effective monitoring tool for environmental health; 3) There exists a high potential to discover biomarkers for environmental changes and/or disturbances.

Following on from this work, we aim to continue to monitor the microbiota in sediment along the current transects, with potential to expand to additional geographic locations. We hope to expand our understanding of microbial communities and interactions, both intra- and inter-kingdom and across different environments and hosts.

MKMRS diver surveying at 45 m depth on a rocky bottom near Nahariya

Biodiversity of the rocky reef

Our long-term study includes different rocky habitats across the Israeli coastline within protected areas (MPAs) and outside MPAs, from Achziv to Ashkelon (up to 45 m depth). At each site, we sampled depths

at set intervals, depending on the rocky reef distribution. Since the reef's ecosystem is dynamic, we decided to sample across two seasons: in spring and autumn to ensure optimal diving and surveying conditions. This work has led to the widening of the protected area at Evtach to include the rocky reef, and establishment of science-based fishing regulations, as well as documenting alien species' establishment in the Israeli coastal rocky reef.

We found marked seasonal differences in the number of fish from all species at all sites. In autumn, there are more fish than in spring, notably large schools of *Chromis chromis* (damselfish) and the invasive herbivore *Siganus rivulatus (rabbitfish)*. Another phenomenon we witnessed was the formation of an invasive population of *Torquigener flavimaculosus* (yellow-spotted pufferfish), and *Pterois miles* (devil firefish) presumed to come from the Red Sea via the Suez Canal. The study within the Achziv reserve has shown us that the shallower depths are a nursery for the predator fish *Epinephelus marginatus*, whereas the larger-sized fish reside in the deeper parts of the reserve.

The rocky reef is mostly covered by turf algae, followed by unidentified branching bryozoa and, to a lesser extent, mixed canopy algae and invertebrates. There was a gradual decrease in algal cover since 2015, and an increase in invertebrate coverage. The algal and invertebrate communities at the 45 m depth sites are richer and more even than the 10 and 25 m sites.

Apex predators

The Marine Apex Predators lab (MAP) focuses on the biology and spatial ecology of pelagic megafauna in the Eastern Mediterranean Sea. MAPare the only laboratory with this scope of focus in Israel and in the Eastern Mediterranean Sea. The gaps of knowledge for these species are enormous, and so we have decided to focus first on the apex predators that we are able to research and access easily, such as the Dusky sharks (*Carcharhinus obscurus*) and Sandbar sharks (*Carcharhinus plumbeus*). Both sharks aggregate in the warm-water effluent of the Orot Rabin power station in Hadera, Israel. In terms of marine mammals, we focus on the stable, coastal populations of the two dolphin species: the Bottlenose dolphin (*Tursiops truncatus*) and Common dolphin (*Delphinus delphis*). In the semi-offshore pelagic waters, we have focused on the Bluefin tuna (*Thunnus thynnus*), the most commercially-valuable fish in the world, which comes to reproduce every spring in the Eastern Mediterranean Sea. Finally, we study the Blackchin Guitarfish (*Glaucostegus cemiculus*) as a soft-bottom flagship species with nursery grounds in Ma'agan Michael and the Evtach MPA.

We employ unmanned aerial vehicles (UAVs), acoustic telemetry, mark-release protocols, underwater observations, and cutting-edge technologies for our biological analyses. Our main research focus includes the abundance, habitat-use and behaviour of elasmobranchs (sharks and rays), teleosts (e.g., tuna and swordfish), andvarious marine mammals in the open sea and at aggregation hotspots (Hadera and Ashkelon power plants, Ashdod open sea fish farm). In addition, the MAPis collaborating with other partners on the development of remote sensing algorithms which allow for fishery-independent sampling of the species under study. So far, 34 females and 1 male of the species *Carcharhinus obscurus,* as well as 19 males and 1 female of *Carcharhinus plumbeus* were measured, tagged and released. These species are commonly referred to as dusky and sandbar sharks and are listed by the IUCN as "Data Deficient" and "Endangered" in the Eastern Mediterranean Sea, respectively. We have recorded three recaptured sharks over consecutive seasons (results not shown). We have also conducted genetic analyses to support the morphological identification of the two above-mentioned species, and reference ranges for blood have been established for a myriad of biochemical parameters. Preliminary data suggest extended residency periods at human-altered habitats and within-season site fidelity to the Orot Rabin power and desalination facility in Hadera

A female dusky shark in the warm waters of the Hadera power plant

In 2016, the MKMRS initiated a detailed study of the bluefin tuna (BFT, *Thunnus thynnus*) population in the Eastern Mediterranean by collaborating with local commercial tuna fishers and the Block Laboratory (Hopkins Marine Station) of Stanford University, USA. BFT is a species of great value due to its biological importance as apex predator. However, they are routinely targeted and fished across the globe due to their high commercial value. A first satellite tagging attempt was conducted off the coast of Israel in 2017, in cooperation with local fishermen from Ashdod.

In total, 89 landed BFT were sampled during three fishing seasons. Genetic indicators confirmed that samples thought to be BFT were indeed *Thunnus thynnus*. In addition, 9 of the sampled fish (two in 2017; two in 2018; five in 2020) were marked with a satellite archival tag and were released back into the sea. One BFT remained in the Eastern Mediterranean Sea for a few weeks before heading to the

central Mediterranean near Malta. An additional tag detached and floated to the surface before the designated time in the vicinity of the Libyan coast (indicating possible capture). Another PSAT tag completed its programmed deployment time, and showed that the fish had travelled to the western basin near Barcelona, Spain and the tag detached on time near Rome, Italy. One more fish, which was tagged in 2019, swam to the western Mediterranean basin and marked a record-breaking monitoring period of 110 days. Other tagged fish revealed a strong Aegean occupancy.

A juvenile guitarfish *(Glaucostegus cemiculus)* along the intertidal zone near Kibbutz Ma'agan Michael (credit: Dr Aviad Scheinin)

In the region of Kibbutz Ma'agan Michael, neonate guitarfish (*Glaucostegus cemiculus)* exhibit uncommon behaviour. They appear to swim onto the beach for short periods before returning to sea. Every year since 2016, MKMRS students and staff have observed, captured, marked, and released hundreds of guitarfish between the end of August and the end of November. In December, there was a marked decline in abundance. Growth curves have been created, and this data has enabled us to characterize the shoreline of Kibbutz Ma'agan Michael as a nursery ground for *Glaucostegus cemiculus*.

Marine pathology program

Studying long-term abundance and prevalence of microbial pathogens in the marine environment is an emerging research field. Marine pathogens abundance is influenced by climate change, may accumulate in commercially important fish and shellfish and have a dramatic effect on mariculture. Some of these pathogens may pass to higher trophic levels such as marine mammals and possess a severe risk also to human health. Fish samples have been obtained from research trawling in the North and South twice a year and from Accre and Jaffa fish markets, to cover main sources of fish. Since the establishment of the marine pathology program in 2016, we have gained preliminary data on the

prevalence of 5 marine pathogens in 400 fish individuals. We have defined protocols for our routine long-term study of pathogens in wild fish populations. During the last six years we evaluated different protocols as wild marine fish pathogens a new monitoring field, therefore we are l continually improving our methodology. Our current system is simultaneous identification of three potential fish bacterial pathogens: *Vibrio harvei, Photobacterium damselae* and *Streptococcus iniae*. In addition, we are performing a pathogen screening program in cetaceans, which led to the first EMS report of *Streptococcus agalactiae* and *Toxoplasma gondii* infection in stranded marine mammals. Lastly, we have published updated values for heavy metal accumulation in various wild fish and shellfish. Four M.Sc. students were involved in the method evaluation process.

Sampling tissue from *Upeneus moluccensis*

The students involved in the Morris Khan Marine Research Station's research so far include:

Tal Elmaliach M.Sc. (Bluefin tuna)

Berzak Ran M.Sc. (Fish Pathogens)

Yael Regev M.Sc. (Fish Pathogens)

Barak Azrieli M.Sc. (Guitarfish nursery grounds)

Tal Ben-Ezra M.Sc. (Nutrient dynamics)

Peleg Itai M.Sc. (Fish Pathogens)

Debra Ramon M.Sc. (Microplastics in the coastal ecosystem)

Yaly Mevorach M.Sc. (Coastal dolphin's population dynamics)

Ori Galili M.Sc. (Coastal dolphins' habitat modelling)

Tal Melonak M.Sc. (Shark and sea turtle haematology analysis)

Rebecca Valani M.Sc. (e-DNA)

Tal Kedem-Zvi M.Sc. (Sediment microbiome)

Adi Barash Ph.D. (Shark populations)

Leigh Livne (née Kroeger) Ph.D. (Natural Resource Management)

Stephane Martinez Ph.D. (Rocky reef food web analysis with stable isotopes)

Ziv Zemach Shamir Ph.D. (Shark behaviour)

Current students are:

Eyal Bigal Ph.D. (Shark population and top predator method development)

Yotam Zuriel Ph.D. (Marine mammal acoustics)

Debbie Ramon Ph.D. (Heavy metals and microplastics in the coastal ecosystem)

Rafael Yavetz Ph.D. (Bivalve metabolism and microbiome)

Ram Ritter Ph.D. (Fish pathogens in aquaculture)

Rami Tzadok Ph.D. (Immigrant fish species ecology)

Tal Ben Ezra Ph.D. (Nutrient limitation in the East Mediterranean)

Ole Johannes Ringnander Soerenson Ph.D. (Seascape fish ecology of the rocky reef)

Alon Blachinski M.Sc (Role of urea in nutrient limitation)

Eitan Newman M.Sc (Heavy metals in tuna blood)

Goni Bergman M.Sc (Shark microbiome)

Kim Kibo M.Sc (Dolphin mother-pup relations)

Lior Shimon M.Sc. student (Gastropod microbiome)

Michal Avital M.Sc. (Zooplankton community analysis with stable isotopes)

Vanessa Bachmann M.Sc. (Sea turtle microbiome and pathogens)

Ximena Dubinsky Velasquez Ph.D. (Anthropogenic influence on copepod microbiome)

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Eyal Miller, Dr Daniel Sher, Tal Ben-Ezra, Prof Danny Golani, Orit Schwartz, Yair Yam and Putzker Diving

Club, Dr Yotam Popowitz, Prof. Yoni Belmaker, Prof. Menachem Goren, Prof. Maoz Fine, Dr Noga Sokolover, Dr Noa Shenkar, Omri Burnstein, Dr Eran Brockowitz, Eran Rozen, Dr Sigal Shefer, Revital Ben David Zaslo, Dr Shevi Rotman, Roni Sade, Dr Ruthy Yahel, Sharon Liper, Tamar Feldstein-Farkash, Assaf Shwartz, James Lindholm, Dr Yair Suari, and Rami Tsadok.

We would like to thank the Kimron Veterinary Institute for their solid collaboration with us.

Many thanks to the researchers and staff of the Steinhardt Museum of Natural History.

The Achziv and Nahariya December 2015 surveys have been carried out with the assistance of the Israel Nature and Parks Authority.

We apologize to whoever was erroneously left out of this list.

To be quoted as: Krom, M. D., Tsemel, A., Scheinin, A., Nativ, H., Morick, D., Meron, D., Bigal, E. Shemesh, E. Azrieli, B. Aharonovich, D., Zuriel, Y., Livne, L., Einbinder, S., Tchernov, D. (2022). Report for activity years 2015-2021 |Seasons 1-10. Morris Khan Marine Research Station, Sdot Yam.

Index

A. Introduction

The Eastern Mediterranean Sea (EMS) in general, and the Israeli coastal shelf in particular, is unusual in a number of important ways. The EMS is a semi-enclosed sea with large inputs of externally supplied nutrients (nitrate and phosphate). These are the fertilizers that enable marine plant growth, yet the EMS has the lowest level of concentration of any ocean in the world (Krom & Suari, 2015, Powley et al., 2017). The EMS is also unusual because the growth-limiting nutrient is phosphorus – which is present in very low concentrations in the Levantine Basin (Krom et al., 1991).

The Sahara Desert is the EMS' terrestrial counterpart. The hot, dry summers in our region cause the surface waters to increase in temperature and salinity. This results in a highly unusual anti-estuarine circulation, which brings nutrient-depleted waters into the EMS through the Straits of Sicily and causes the nutrient-rich waters to be jetted out via the deeper outflowing waters. This means there are very low levels of nutrients for algae and plants to grow and an unusual semi-tropical (but very limited) marine ecosystem. It is the reason why the EMS is so blue, and not as green as other seas.

The EMS coastal ecosystem has been subject to fishing pressure, climate change, and pollution, which all have a major effect on the ecosystem. Prior to the completion of the Aswan Dam in 1965, there was a plume of nutrients from the annual Nile flood that were routinely discharged into the Egyptian (and thence) the Isreali coastal shelf, which resulted in a major pulse of marine plant growth. This, in turn, provided food for the local ecosystem (Halim, 1960; Nixon, 2004); this nutrient-laden flood reached as far as Beirut. After the Aswan Dam was completed, the annual nutrient pulse stopped and the primary productivity was drastically reduced (Suari & Brenner, 2015), which had a knock-on effect on the entire ecological system (Nixon, 2004).

Added to this major environmental change is the effect of the largest influx of exotic marine species via the Suez Canal in the world (Galil, 2008). Since the Red Sea and EMS are connected via the Suez Canal, (which is in the process of enlargement), Red Sea organisms are flooding into the Israeli coastal shelf and taking over shallow water habitats (Edelist et al., 2013). Since 2008, more than 68 fish species have migrated into the EMS (Golani et al., 2008; Fricke et al, 2017) some of which have established large populations. As a result, more than 70% of the fish caught in Mediterranean coasts of Egypt are Red Sea species (Edelist, 2012). These species are better adapted to the semi-tropical conditions in our coastal region than the endemic Mediterranean species which originated in the temperate Atlantic sea. The effect of climate change on the system can already be seen, as the surface waters are becoming systematically warmer (Gertman et al., 2016) and more acidic (Bialik & Sisma Ventura, 2016). Finally, populations of fish and marine apex predators throughout the world have dramatically declined over the past seven decades. Intense exploitation and overfishing have left many fish stocks depleted, resulting in the dire consequences expressed in the lower trophic levels and felt throughout the ecosystem (Letessier et al., 2017). In the past few decades, there has been a worldwide increase in reports of diseases affecting marine organisms of different taxa. Climate warming can also increase pathogen development and their survival rates, disease transmission, and host susceptibility, but there is a lack of baseline data regarding pathogenic agents' prevalence in wild fish and other marine animal populations.

Both bottom-up and top-down research is carried out at the Morris Khan Marine Research Station in Kibbutz Sdot-Yam—bottom-up through the nutrient measurements and their control of microbial growth at the base of the food web, and top-down through the marine apex predators. Marine apex predators play a crucial role in maintaining the integrity of ecosystems in both function and structure by virtue of their position at the top of the food web (Letessier et al., 2015). This group of predators include coastal and pelagic species of marine mammals, as well as bony and cartilaginous fishes, such as sharks and tuna. In the EMS, knowledge gaps relating to the abundance of highly migratory, pelagic megafauna translate into inadequate scientific data to inform policies with regards to apex predators.

The Morris Khan Marine Research Station (MKMRS) initiated the long-term study of biodiversity in 2014 in several locations along the Israeli coast. The goals of the activities are:

- To expand upon the available scientific data set to enable the progress of ecological research in the Israeli coastal zone.
- To ensure the data is accessible to enable better planning, management and policy making, as well as for other interested parties.
- To carry out real-time identification of changes in status of ecological systems.

The steering committee who determined the various components of the long-term research conducted by MKMRS include experts from a diverse field of marine research: Prof. Dan Tchernov, Prof. Jonathan Belmaker, Prof. Maoz Fine, and Prof. Hezi Gildor. The Achziv survey requirements were defined by Dr Ruthy Yahel. The program is in cooperation with the Leon H. Charney School of Marine Sciences, of the University of Haifa.

B. Data collected and sampling sites

Table B.1: Data collected in the MKMRS, data period of this report and additional details, as of June 2021

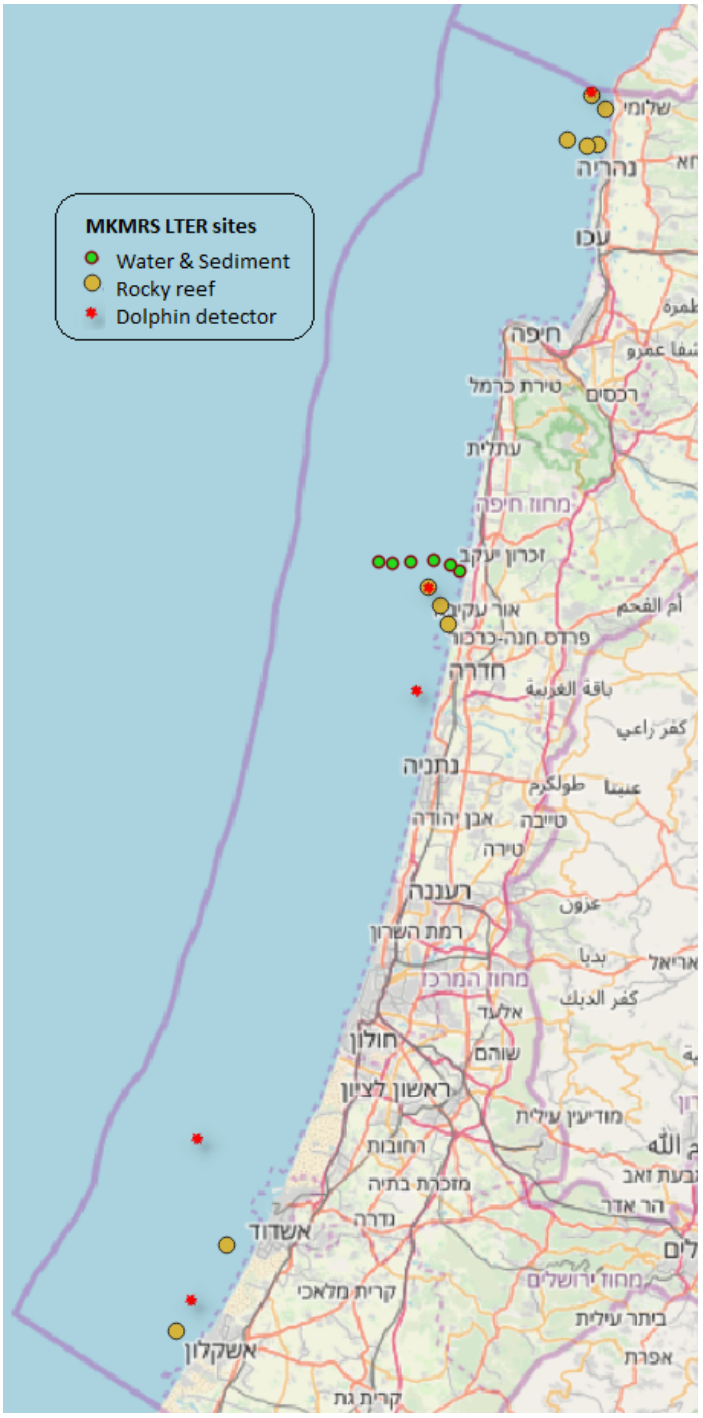
Data Type	Depth	Data period covered in this report	Data collection period	Site Depth	Sites
Salinity, temperature, chlorophyll fluorescence	20	Entire data	Jan-March & July-Sep 2016, March-July & Nov 2017-Feb 2018 July-2018 –Jan 2019 March-June 2019	32 m	Achziv
Salinity, temperature, chlorophyll fluorescence, Inorganic Nutrient content, Dissolved organic and particulate N and P	Profile data	Dec 2017 - Feb 2021	Dec 2017 - June 2021	10, 25, 45, 70, 100m	Ma'agan Michael transect
Chlorophyll-a (extracted)	Surface waters	None	April 2016 – June 2021	10, 25,45 m	Achziv, Nahariya, Sdot Yam,
	Surface waters	None	May 2017-June 2021	30 m	Ashdod Ashkelon
Soft-bottom microbial communities	10, 25, 45, 100	Entire data	July 2017 - June 2021	10, 25, 45, 100 m	Sdot Yam
Bacterial counts	5 m and bottom depth	Entire data	April-May 2015- June 2021	10, 25, 45 m	Achziv, Nahariya, Sdot Yam, Ashdod Ashkelon
Zooplankton	5 m	July 2019	July 2019 - June 2021	25, 45, 100 m	Sdot Yam
Algae and invertebrates	Bottom Depth	April-May 2015-	April-May 2015- June 2021	10, 25, 45 m	Achziv, Nahariya, Sdot Yam
Fish	Bottom Depth		May 2017-June 2021	30 m	Ashdod, Ashkelon
Coastal shark species	Surface waters	November 2016 - May 2021	November 2016 - June 2021	1-8 m	Hadera

Table B.1: Data collected in the MKMRS, data period of this report and additional details, as of June 2021 - continued

Data Type	Depth	Data period covered in this report	Data collection period	Site Depth	Sites
Stranded Marine Mammals via a bank of tissue samples		None	Samples have been organized from the former University of Haifa (IMMRAC) tissue bank (1993-June 2021)	-	Israeli Mediterranean coast
Dolphins: presence/absence		2018-2020	December 2018-June 2021	25-45	Nahariya, Dor, Michmoret, Hof Hasharon, Ashdod, Ashkelon
Rays	0–2 m	2016-2020	August 2016–June 2021	0–2	Ma'agan Michael, Evtach MPA
Marine Pathology	20-80 m	May-June 2021	May 2016-June 2021	various	Ashdod (trawler) Annual wild fish collection. 150 fish from 5 different species were collected as planned.

The majority of the long-term ecological research (LTER) activities are conducted at two sites, Achziv and Sdot Yam. Achziv and Sdot Yam are surveyed for fish, algae and invertebrates at three depths: 10, 25 and 45 m. At these sites, water is being collected for bacterial counts, and chlorophyll content. The same data collection is also carried out at the sites of Nahariya (45 m), Ashdod (30 m) and Ashkelon (30 m). The data collection is conducted during May-June and October-December, when the sea is calm for longer periods, and visibility is high. The sites are re-located using coordinates and a GPS device, but the transects are randomly set.

A stationary CTD (conductivity, temperature, and depth device) was deployed in January 2016 at the 20 m isobath, along the 32 m contour line in Achziv. The CTD recorded temperature, salinity, fluorescence, and was lost to sea after June 2019, possibly due to trawler activity.



Map of MKMRS sampling sites and parameters collected within the Israeli territorial waters

A transect offshore of Ma'agan Michael was added to the MKMRS long-term ecological activities in December 2017, sampling water for CTD profiles, cell counts, chlorophyll, nutrients in the water and in the dissolved and particulate fractions, and sediment for archaea and eukaryotes. This is routinely carried out at five depth stations from 10 to 100 m. During 2020 we added an open sea station (850 m bottom depth) to this program.

Fish were collected to analyze pathogens from trawl tows in three areas: North (Akko), Center (Jaffa) and South (Ashdod).

Dolphin acoustic presence has been monitored at various sites along the Israeli coastline since 2016, and we are also tagging and monitoring shark presence though our catch-and-release programme in Hadera. In the south, apex predators are being recorded in Ashdod open-water fish cages and on pelagic longlines set by local fishermen. Guitarfish have been consistently surveyed at the Ma'agan Michael nursery grounds since 2016. The full dissemination of parameters and collection is described in Table B.1.

1. Oceanographic Parameters

Scientific supervision: Dr Yair Suari

Table 1.1. Oceanographic parameters collected using the Seabird (SBE) 16 CTD , which was suspended at 20 m depth along the 32 m contour near Achziv near the shipwreck Nitzan.

Data Collected	Planned data	Problems encountered	Main results	Suggested improvements
Temperature, Salinity, Fluorescence, Turbidity, Oxygen	*in situ* CTD time-series at Achziv	CTD was lost after June 2019, possibly due to trawler activity	Significant increase in salinity in 2018 relative to 2016 and 2017.	Two new sensors need to be purchased to facilitate a continuous time-series with one under maintenance and one currently sampling. We recommend sensors with shorter maintenance time.

In an environment affected by climate change and anthropogenic activities, it is important to monitor environmental parameters which may also then be used to explain specific ecosystem processes.

The data presented here is available to download at:

https://med-lter.haifa.ac.il/index.php/en/data-base under Marine Chemistry/CTD station Nitzan

Methods

A Seabird 16 plus CTD measuring salinity, temperature, turbidity, and fluorescence (a proxy measure for chlorophyll-a) and was installed in January 2016 at a depth of 20 m at the 32 m contour line near Achziv.

Results

The data collection (monthly) of the Nitzan CTD are depicted in Table 1.2.

Table 1.2 : Data collection of the Nitzan CTD, which was suspended at 20 m depth along the 32 m contour near Achziv.

Year	*Jan*	*Feb*	*Mar*	*Apr*	*May*	*Jun*	*Jul*	*Aug*	*Sept*	*Oct*	*Nov*	*Dec*
2016	*X*	*X*					*X*	*X*	*X*	*X*		*X*
2017	*X*	*X*		X	X	*X*	*X*	*X*			*X*	*X*
2018	*X*	*Overseas maintenance*					*X*	*X*	*X*	*X*	*X*	*X*
2019	*X*		*X*	*X*	*X*	*X*	CTD lost					

The annual data is depicted in Figures 1.1 and 1.2. There is variability in the data, and is too short and fragmented to be able to assess long-term processes.

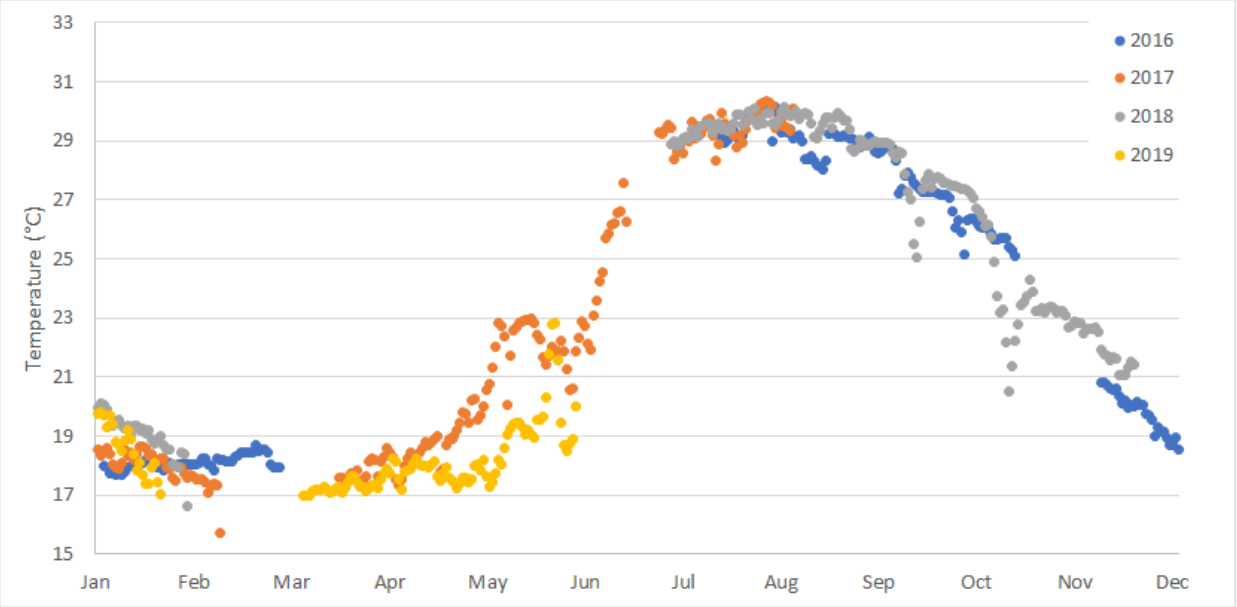


Figure 1.1: Daily average of the seawater temperature as recorded by the Nizan CTD at a depth of 20 m near Achziv, January 2016 - May 2019.

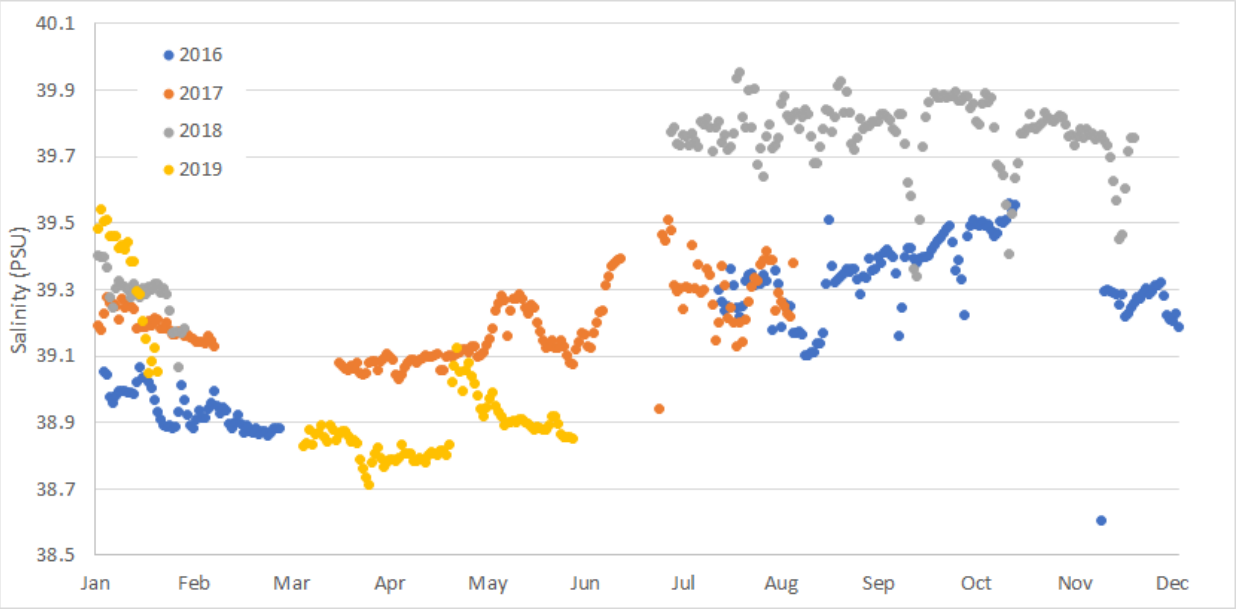


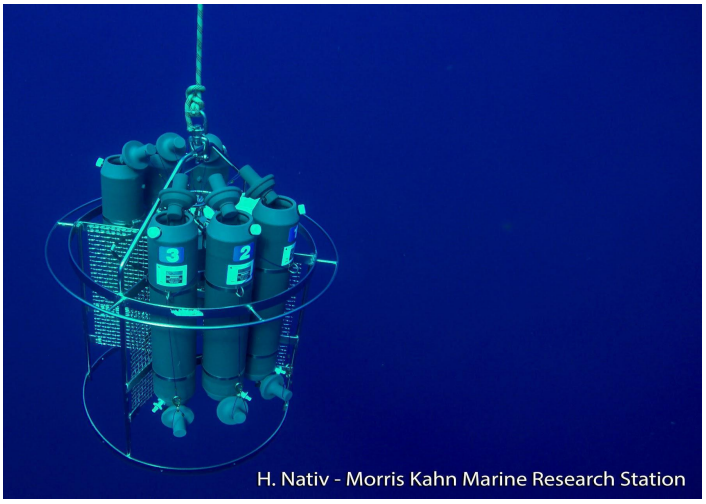
Figure 1.2: Daily average of the seawater salinity as recorded by the Nizan CTD at a depth of 20 m near Achziv, January 2016 - May 2019.

2. Marine biogeochemistry

Scientific supervision: Prof. Michael D. Krom

Table 2.1: Summary of activity for the marine biogeochemistry lab, 2015-2021

Data Collected	Planned data	Problems encountered	Main results	Suggested improvements
1. Nutrient content in seawater: nitrate + nitrite, silicate, ammonium, soluble reactive phosphate 2. Dissolved organic phosphorus and nitrogen, urea, chlorophyll, and particulate matter N & P 3. CTD salinity, temperature, depth, fluorescence	1. CTD data and seawater at the Sdot Yam transect stations 2. Nutrient measurements at the LTER rocky sites 3. Nutrient preservation experiments	LTER rocky reef samples taken using syringes while diving resulting in nitrate & NH4 pollution of the samples	1. Nutrient concentrations measured above the shelf and in the offshore are low 2. Ma'agan Michael transect results suggest an input of nutrients from upwelling mostly, as well as lesser input from land and from the atmosphere 3. Preservation affects dissolved phosphorus and nitrogen seawater content	Need to collaborate with a physical oceanographer for better understanding of oceanic physical processes



SBE55 mini-rosette with CTD used for the first time on the Sdot Yam cross-shelf water transect: an in-water perspective

The Morris Kahn Marine Research Station is carrying out long-term measurements to determine the effect of environmental and climate change on the EMS coastal and offshore ecosystems. The LTER rocky reef nutrient data and basic analyses are also

available at: Morris Kahn Marine Research Station - Water chemistry. Additional data may be available upon request.

Methods

The hyper-oligotrophic conditions in the EMS are challenging. It took our researchers the better part of three years to develop sampling protocols which preserved the nutrient concentrations from the field to the lab and to develop analytical procedures to reach the highest possible detection ability for nutrients in seawater. Our results from this work are presented in Table 2.1 below.

A cross-shelf water transect was sampled off the shores of Ma'agan Michael using a SBE55 mini-rosette at depths of 10, 25, 45, and 100 m for dissolved nutrients (phosphate, nitrate, ammonia and silicic acid), dissolved organic phosphorus (DOP), and chlorophyll, for lab analysis.. We also recorded salinity, temperature, depth, chlorophyll fluorometry, and turbidity data from the CTD. From February 2018 onwards, we added another station at the 70 m contour line. All samples are filtered immediately and then stored at 4 ℃ until they can be analyzed within 12-18h of collection. Filtered frozen samples are used as backups if needed. The procedure of using unfiltered frozen samples, which was the previous method used for sampling on the ICS, was proved to be less acceptable.

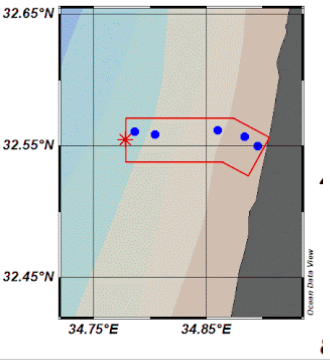
The analysis was carried out on a SEAL Analytical 3 Autoanalyzer (AA3) using specific colorimetric methods developed for nitrate+nitrite, silicate, soluble reactive phosphorus (SRP), (SEAL Analytical Methods a, b, c), and a fluorometric method for ammonium (SEAL Analytical Method, d) using a Jasco FP 2020 plus fluorometer. The SRP method detection limit was pushed lower using a World Precision Instrument (WPI) detection cell 1 m long (LWCC-210). The sensitivity and limit of detection for the described methods are in the following table (Table 2.1). The methods used in the lab are now routine (SRP, ammonia, and silicate). The nitrate method will be changed to an ultra-low sensitivity method with a new long flow cell (LWCC-210, expected precision of 2 nM).

Table 2.1: Typical range of nutrients in seawater, precision and limit of detection of methods used for the results presented in this report.

Determinant	***Typical Range (nM)***	***LOD [nM]***	***Precision [nM]***	***Comments***
SRP	*0-125*	*2*	*6*	*Using LWCC*
TOxN	*0-1000*	*9*	*34*	
Silicic acid	*0-2043*	*8*	*11*	
Ammonium	*0–460*	*5*	*5*	*Fluorometer, care was taken to minimize contamination*
Dissolved organic Phosphate	*0–200*	*5*	*2*	*Using LWCC*

Table 2.2 Sdot Yam transect station locations

Ma'agan Michael transect stations	lat	long
SY_W10	32.550	34.896
SY_W25	32.557	34.884
SY_W45	32.562	34.860
SY_W70	32.559	34.804
SY_W100	32.561	34.786



Results

From the initial two years of sampling along the Ma'agan Michael transect (MMT), we have observed that the nutrient concentrations are generally very low, but there are complex patterns, the sources and sinks of which are yet to be clarified. Exemplar profiles are presented below from two sampling dates. On February 20, 2018, we observed a surface plume of Soluble reactive phosphorus (SRP, phosphate)

and nitrate+nitrite (N+N), likely of terrestrial origin. On the same cruise, there was evidence of upwelling (increased N+N) of the offshore waters at 80-100 m depth of the 100 m station (Fig 1.1).

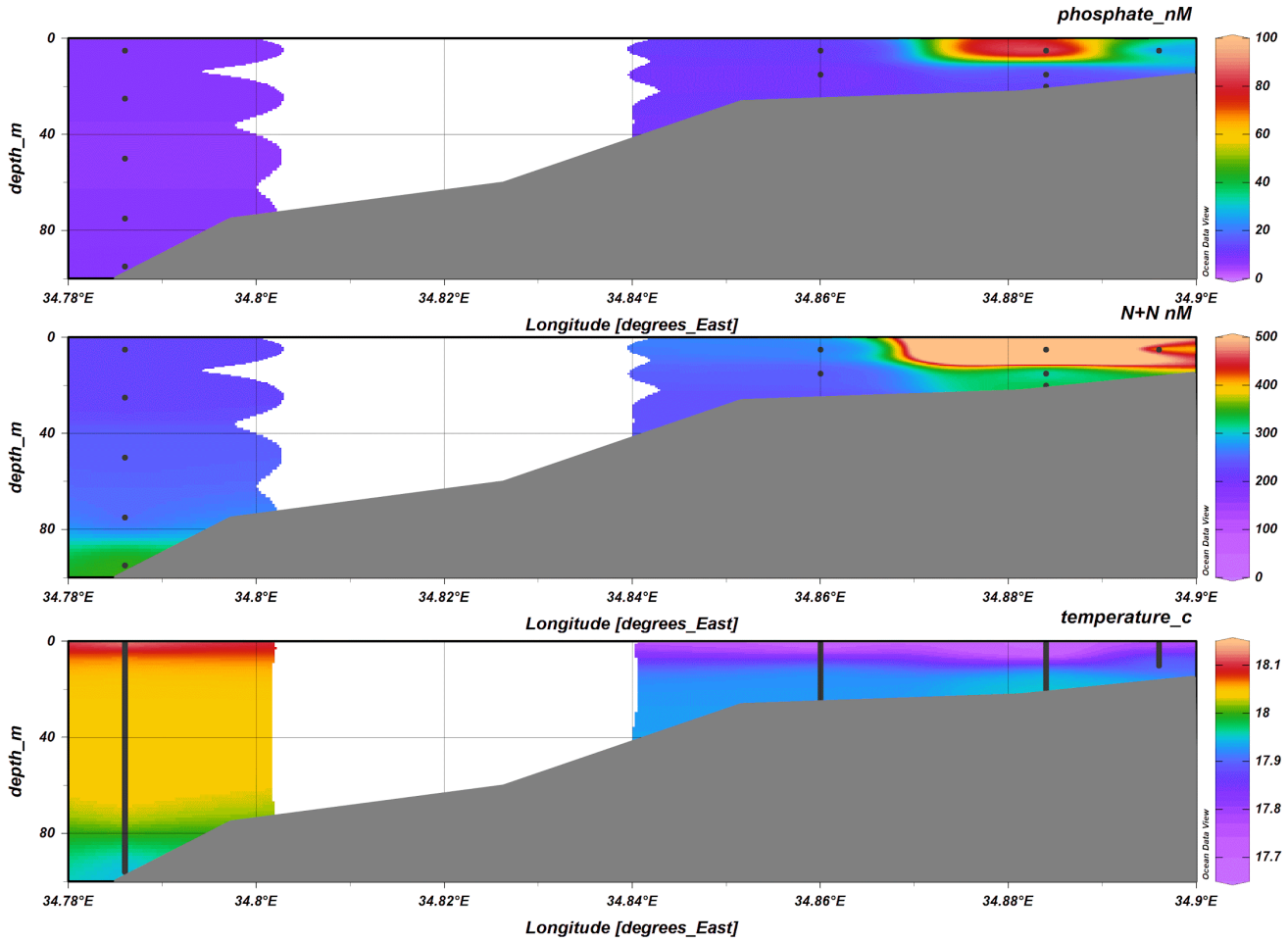


Figure 1.1: Phosphate, N+N, and temperature along the Ma'agan Michael transect on February 20, 2018. Each black dot is a datum; weighted average gridding (nutrient scaling: X scale: 70; y scale: 60 permille).

From the May 2018 transect (Figure 1.2), we observed a strong stratification of Nitrate (N+N), both from atmospheric and upwelling sources.

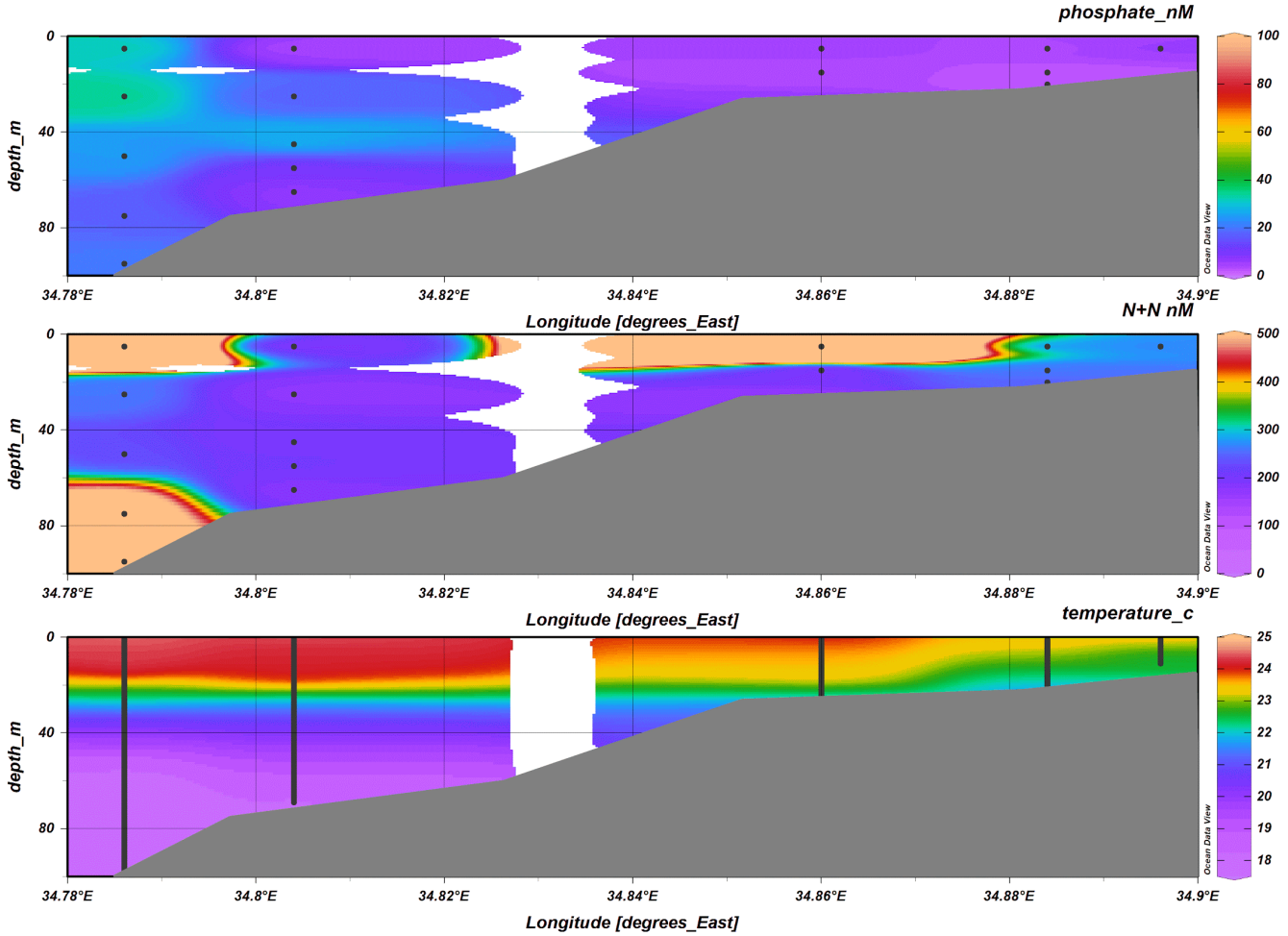


Figure 1.2: Measurements of phosphate, N+N and seawater temperature recorded along the Ma'agan Michael transect on May 29, 2018. Each black dot represents a datum; weighted average gridding (X scale: 70; y scale: 60 permille).

Looking at ourtime series data (Figure 1.3) 100 m depth stations over time, we observed that the Israeli coastal shelf had very low concentrations of dissolved nutrients throughout the study, generally at similar concentrations that are reported in ultra-oligotrophic pelagic waters. Here, we observed a significant input of nutrients from upwelling up to 80 m depth. We are currently developing a total N budget for the Israeli coastal shelf (ICS), which shows that upwelling is the major source of N to the shelf. This is direct support that nutrient upwelling onto the ICS is an important process and is derived from the observation that there were increased nutrients at the 100 m station of the MMT, but not at 70 m depth. Since the underlying sediment at 70 m and 100 m is similar in grain size and organic

matter content, this implies that the increase in deep water at 100 m was due to advected water from offshore and not from extensive nutrient flux from the underlying sediment.

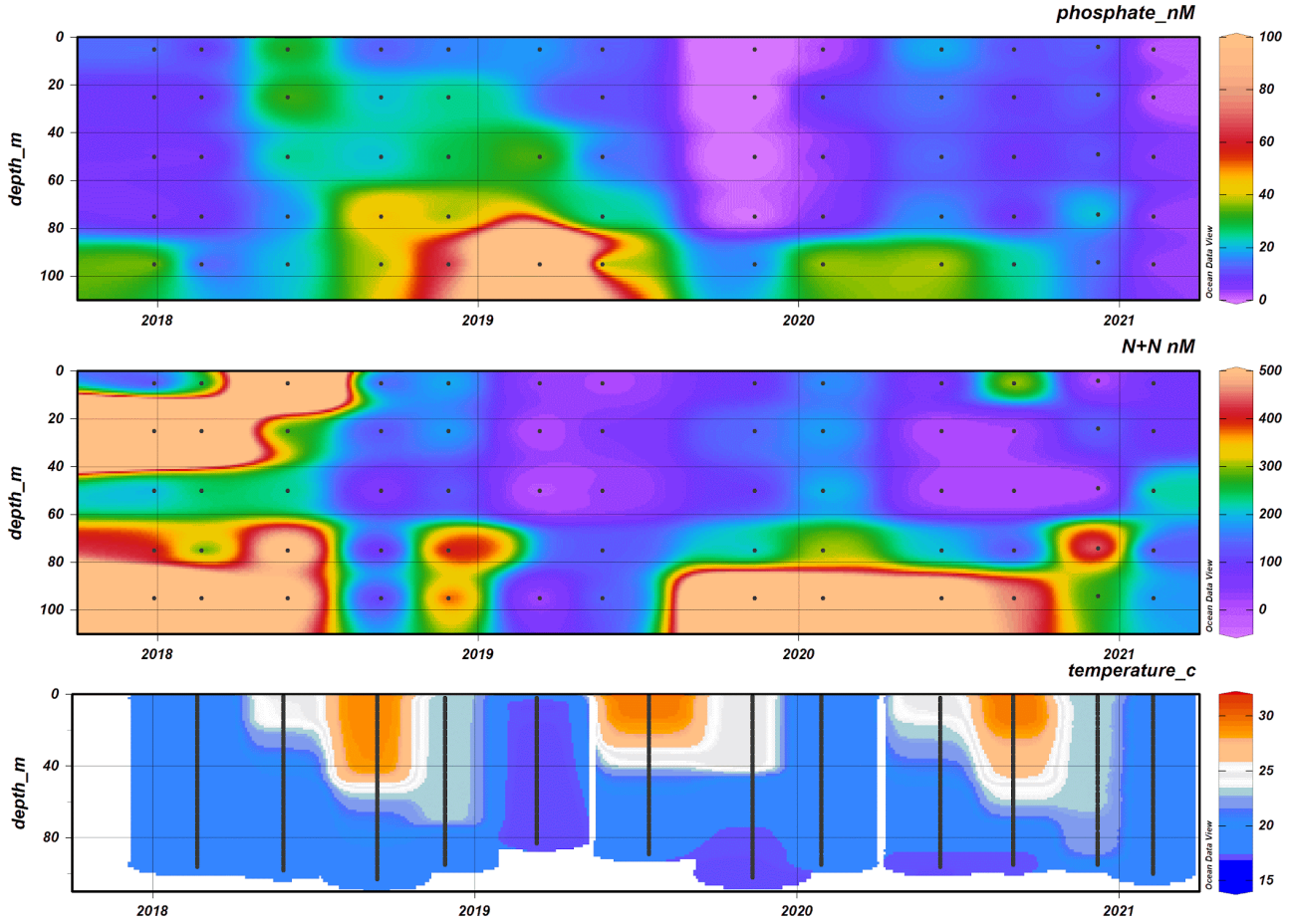


Figure 1.3: Phosphate, N+N and sea water temperature recorded at the 100 m depth station of the Ma'agan Michael transect between December 2017 and February 2021. Each black dot represents a datum; weighted average gridding (X scale: 70; y scale: 60 permille).

Some preliminary insights gained include:

- **The shelf behaves similarly to offshore waters; it has similar nutrient concentrations, low chlorophyll and nutrient dynamics. The water column is depleted in SRP all year, has higher N+N in winter and depleted N+N in summer.**

- **There is no obvious nutrient signal coming from the upper sediment layers. Indeed, it is likely the sediments are a sink of nutrients since they are within the photic zone.**

- **The nutrient inputs are dominated by upwelling**

- **The offshore sampling exhibited major annual variation which suggests a possible pattern**

related to climate change impacts; we noted that the shelf has similar variations (results not presented here)**.**

- **We may have missed important patterns since our sampling scheme missed the month of March each year.**

- **On some occasions, there werehigh nutrient concentrations coming off the land into the shallow waters (10 and 25 m).**

These results are consistent with results obtained by Prof. Berman and colleagues during the 1980's and have not been repeated since (Berman et al., 1986). We expect to continue our work using modern technology to develop a better understanding of how this important ecosystem works, how it worked in the past, and its future response to environmental and climate change for enhanced, science-based management decisions.

In addition to this coastal long-term research, the marine biogeochemistry group has carried out the first detailed seasonal nutrient dynamics study at a pelagic (offshore) station at 800 m bottom depth (Ben-Ezra et al., 2021). The study found that there was a seasonal change from high nitrate in winter to low nitrate in summer, with low phosphate year-round. This seems to be a previously unrecognized characteristic of P-depleted ocean systems. We have been studying this station each month during 2018 and 2019, and seasonally since. We have carried out nutrient limitation experiments of both the autotrophic and heterotrophic microbial communities, which show unusual patterns which we expect will be used to characterize P-depleted oceanic systems.

We have also examined the effect of natural climate change in the region and showed that between 6,000 and 12,000 years ago, the EMS was very different, as the deeper waters were entirely depleted in oxygen. In addition, the waters of the Israeli coast developed an oxygen minimum zone related to the increased primary productivity caused by increased flow and nutrient supply from the River Nile. The study suggested that such changes might be used as natural paleo-examples for modern climate change processes (Zirks et al., 2021).

3. Bacteria and phytoplankton in the seawater

Analyzed by Dr Dikla Aharonovich, the Daniel Sher Lab of Marine Chemical Ecology

Table 3.1: Summary of activity for the bacteria and phytoplankton in seawater, 2016-2021

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Bacteria and phytoplankton counts conducted via Flow cytometry	Bacteria and phytoplankton at surface and bottom depth at the rocky reef sites.	None	1. Total cell counts are affected by year, with higher cell counts at almost all sites and depths during spring and summer of 2020.	None
			2. Total cell counts are affected by season and depth: greater cell abundance in spring versus autumn; greater abundance of cells near the surface than near the bottom.	
			3. Phytoplanktonic cell counts follow similar trends, one order of magnitude lower.	

Primary producers compose the base of the food web. They grow by consuming CO_2, light, and nutrients, and are then consumed by zooplankton. However, in the eastern Mediterranean Thingstad et al. (2005) have observed nutrient cycles within the water column in the form of direct consumption of bacteria by zooplankton, before the former decompose and release nutrients into the water. Long-term research on cell counts aims to understand the basis of the producer-recycler communities of the rocky substrata, in order to have these data available for further analysis in the future. This method has been employed since April 2015. LTER bacterial data is also provided at: Morris Kahn Marine Research Station - Water chemistry.

Methods

Three sampling replicates of 2 ml of seawater each are collected using a syringe in surface waters (5 m depth) and also at a bottom depth (25-45 m) for each rocky reef site. Two samples were analyzed and one sample was kept as an option for future analysis, or as backup. The samples are filtered through an 80 µm filter, then fixed with glutaraldehyde (0.125% final concentration), incubated in the dark for 10 minutes, flash frozen in liquid nitrogen and stored at -80 °C. Before analysis, samples were thawed at room temperature and run on a flow cytometer BD FACSCanto™ II Flow Cytometry Analyzer Systems

(BD Biosciences) to determine phytoplankton and total cell counts. Each sample was run twice with 2 µm diameter fluorescent beads (Polysciences, Warminster, PA, USA) as a size and fluorescence standard. In the first run, five types of phytoplankton cells were identified based on their natural auto-fluorescence. Cells were differentiated based on cell chlorophyll (Ex482nm/Em676nm, PerCP channel) and phycoerythrin fluorescence (Ex564nm/Em574nm), and by the size of cell (forward scatter). Before the second run, samples were stained with SYBR Green I (Molecular Probes/ ThermoFisher) detection at Ex494nm/Em520nm (FITC channel). This provided counts of the total bacterial population as well as a distinction between cells with high or low DNA content. The analysis and results are carried out using FlowJo software. Flow rates are iteratively set during each run and the average rate is used for determining the cell count per ml. The phytoplankton are classified to the Genera level (*Prochlorococcus, Synechococcus*), as well as Picoeukaryotes, according to size and autofluorescence of their natural pigments. Marine bacterial cells are typically composed of two populations: high DNA and low DNA (as depicted in Figure 3.1).

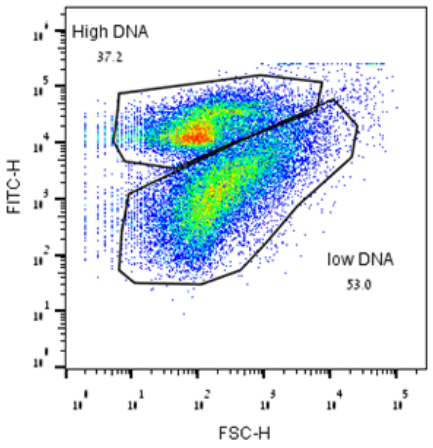


Figure 3.1 FlowJo graphical output of a typical sample of seawater. Each dot on the figure is a cell DNA dyed with SYBRGreen.

The phytoplanktonic population structure is grouped as either high DNA or low DNA according to size (x axis) and fluorescence (y axis), and depicted as a polygon (Figure 3.2).

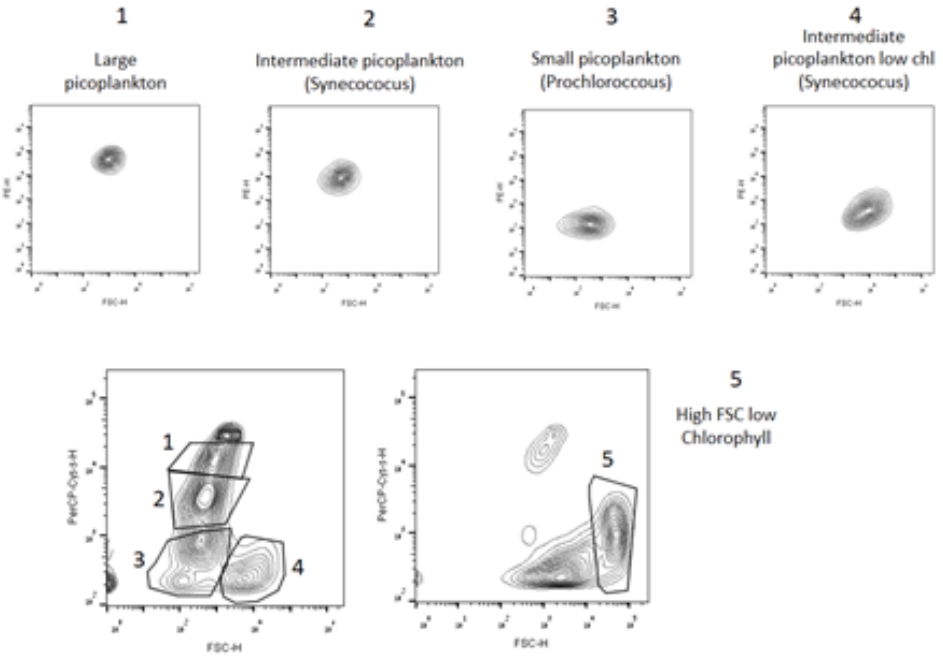


Figure 3.2 Autofluorescence of phytoplankton cell population, grouped as polygons of different sizes and autofluorescence. Community structure is grouped into polygons (marked 1 to 5) of different sizes (x axis) and fluorescence (y axis).

Results

Below we present the total cell count across the different rocky reef sites (Figure 3.3). Cell counts in 2020 were almost twice as high as any other year, in both seasons. Our data also show that surface samples have more cells than bottom samples, and that the cell numbers are higher in the spring versus the autumn (not shown here).

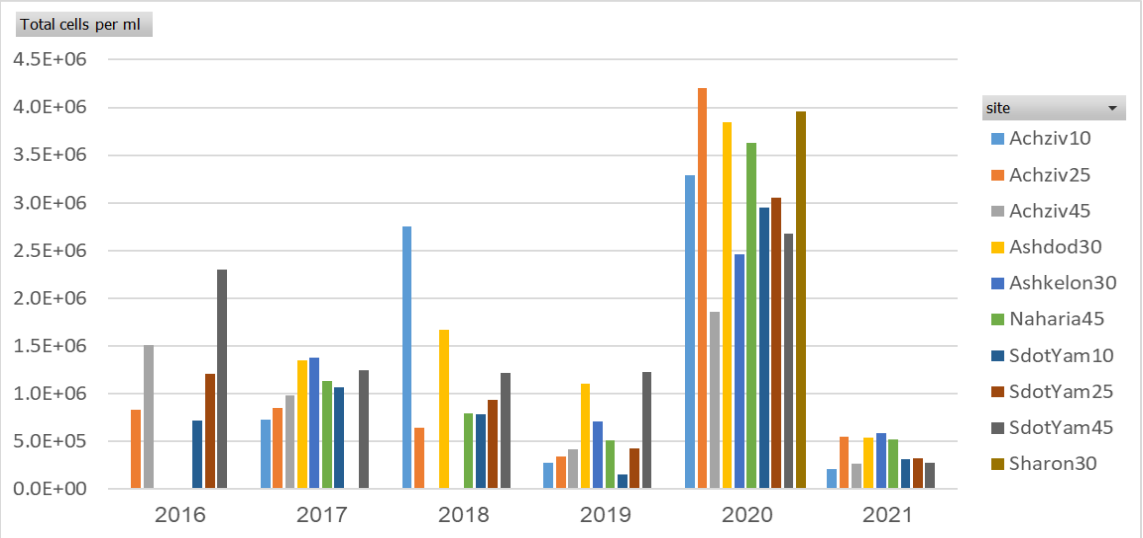


Figure 3.3. Mean number of cells per ml recorded in the rocky reef sites at different sites, bottom depths (10 m, 25 m, 45 m), and years. Surface/bottom and season have been pooled.

Phytoplanktonic cell numbers (Figure 3.4) follow the trend of the overall cell numbers, only one order of magnitude lower.

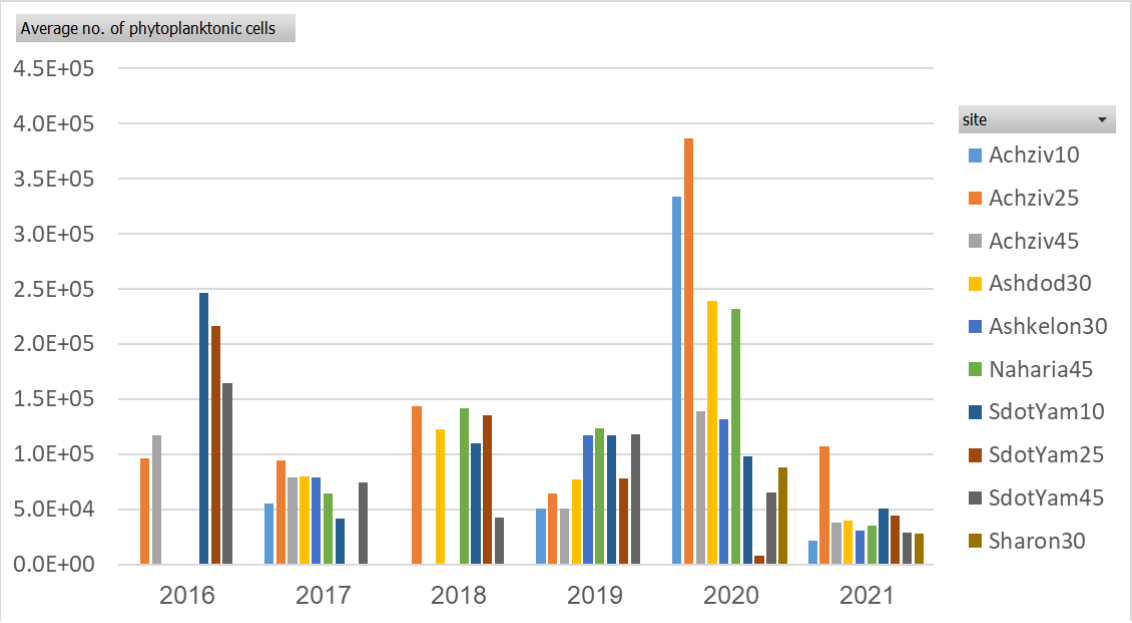


Figure 3.4. Mean number of cells per ml recorded in the rocky reef sites at different sites, bottom depths (10 m, 25 m, 45 m), and years. Surface, bottom and season have been pooled.

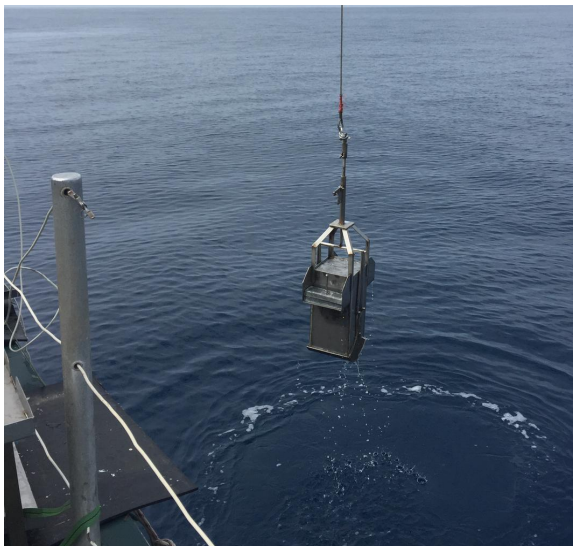
4. Sediment microbial communities

Scientific supervision: Dr Dalit Meron

Table 4.1: Summary of sediment microbial community activity from 2017-2020

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Microbial communities including: bacteria, archaea and eukaryotes.	Sampling twice a year at the Sdot Yam site, along a cross-shore transect with varying depth transect from 10-100 m	Sampling problems encountered with box coring were solved during the year	Significant differences across all parameters examined between samples from different depth core layers and seasons	1. Overlapping the sediment transect with the water chemistry transect. 2. Selecting only relevant slices from each core for further analysis 3. Expanding the sampling sites (both north and south of the ICS)

Oceans cover 70% of the earth while sedimentary habitats cover most of the ocean bottom (ranging from gravel to fine muds), making this the largest ecosystem on our planet in terms of area. Surface sediment bacteria play a significant ecological and biogeochemical role in marine ecosystems due to their high abundance relative to the overlying water column, and they play a key role in the decomposition of organic matter, nutrient and sulfur cycling, and carbon flux (Seymour 2014).

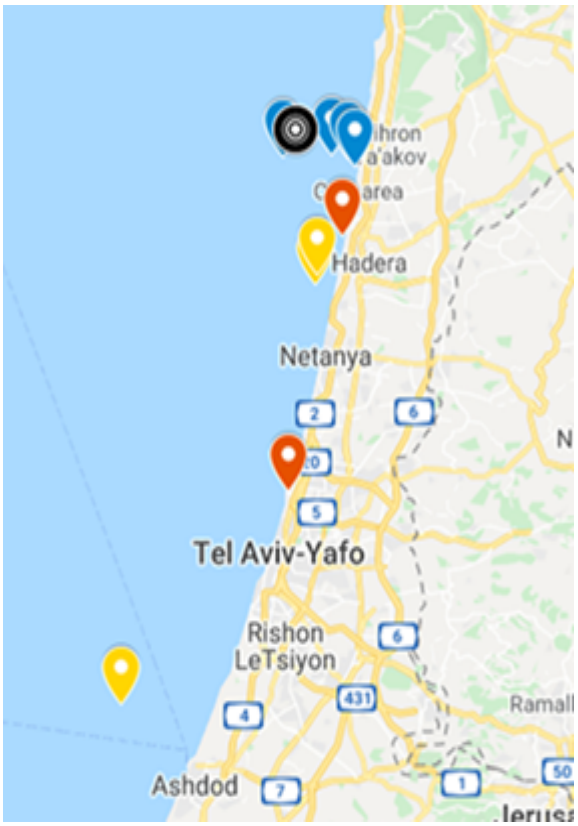


Box corer deployed off the Mediterranean Explorer research vessel

Bacteria can alter sediment chemistry which then determines contaminant bioavailability (e.g. reducing metal bioavailability). As bacteria represent the basal element in the food web, these processes may facilitate the bioaccumulation of metals in higher trophic organisms (Kennish 2002). In addition, metabolites produced by bacteria are also known to increase the rate of mineral dissolution in surrounding sediments (Gadd and Griffiths 1977).

The microbial community is very dynamic and sensitive, and can rapidly change due to environmental changes. Therefore, the characterization of the sediment microbiota may be used as an important tool for assessing environmental health and indicating change to thethe ecological system (Dauer 1993). Considering their importance for ecosystem function and their functionality as an environmental indicator, our knowledge of the bacteria that inhabit the surface sediments is very limited, especially along the Israeli continental shelf (ICS), which is replete with complex human interactions. The LTER sediment microbial community data and basic analysis is provided at: http://med-lter.haifa.ac.il/index.php/data-base/microbiology

Methods



Sampling is carried out twice a year at the Sdot Yam site along four depth transects: 10, 25, 45 and 100 m, and additional sites (Figure 4.1). Two cores from each depth are taken (by diving or by box-corer) and each core is split to 1 cm slices (up to 10 cm). Then, DNA is extracted and analyzed in order to characterize the bacterial, archaeal and eukaryotic communities.

Figure 4.1 Map of sampling sites including theSdot Yam transect (10, 25, 45 and 100 m in blue); "Disturbed sites" are colored red and include Herzliya marina (3 m) and the Hadera power plant (6 m); and "undisturbed tst sites" are colored yellow and include Michmoret (38 m) and Ashdod (70 m). In addition, the position of the Leviathan natural gas platform is indicated in black.

Results

These results are an interim summary of sampling from 2017-2020 (over six seasons) along the Sdot Yam transect (sampling at depths of 10, 25, 45 and 100 m). It is important to note that the deepest sampling point, 100 m, is located about 1 km from the 'Leviathan' natural gas treatment platform (Fig. 4.1).

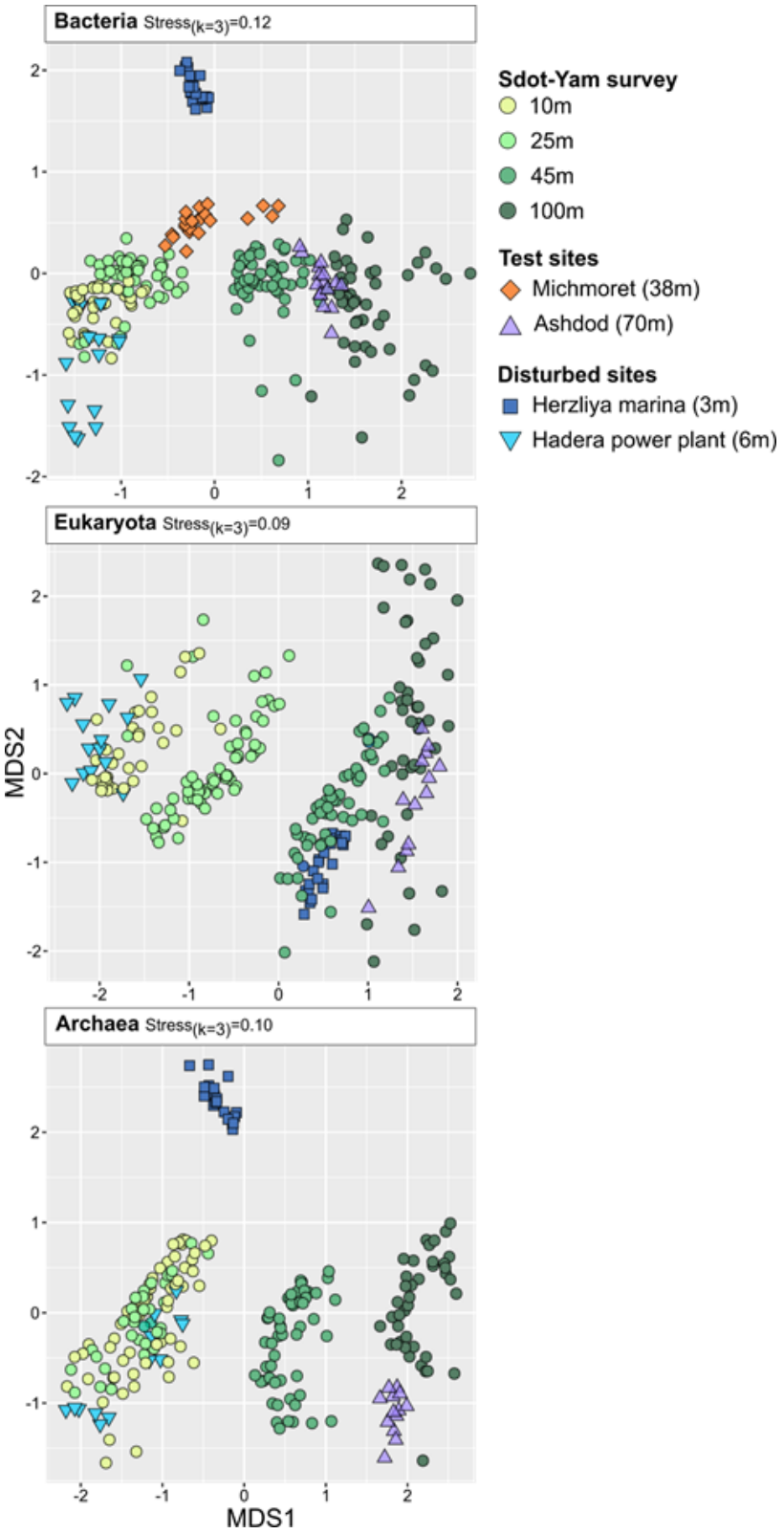


Figure 4.2: High-throughput amplicon sequencing of small subunit rRNA genes illuminates sediment microbiota composition robustness and response to environmental conditions. Sediment samples were collected at Sdot Yam site at 10-100 m bottom depth over six seasons, from 2017 to 2020. Also included were samples from the "undisturbed sites": Michmoret and Ashdod and the"disturbed sites": Herzliya marina and Hadera power plant. Compositions of Bacteria (top), Eukaryota (middle) and Archaea (bottom) were compared by non-metric multidimensional scaling analysis (NMDS) using the Bray-Curtis distance metric.

We started monitoring in February 2017, before the gas platform initiated its production (December 2019). We will continue to sample and monitor this site over the course of the

project and will note possible effects.

*Table 4.2: Effect of bottom depth, core depth and season on sediment microbiota. PERMANOVA factorial test results. R2 values, describing the relative contribution of each factor to variation in microbiota composition are presented. ** denotes a significant effect, P < 0.01.*

	PERMANOVA R^2 value		
Factor	Bacteria	Eukaryota	Archaea
Season	0.027**	0.024**	0.021**
Bottom depth (BD)	**0.168****	**0.067****	**0.1****
Core depth (CD)	0.053**	0.032**	0.037**
Season x BD	0.019**	0.015**	0.02**
Season x CD	0.013	0.01**	0.005
BD x CD	0.026**	0.016**	0.015**
Season x BD x CD	0.009	0.009**	0.006

The sediment microbiota across the three kingdoms showed stability of composition over three years of sampling (Fig. 4.2). Bottom depth was the main contributor to variation in microbiota composition (7%-17% of variance), followed by core depth (3%-5% of variance) and season (2%-3% of variance (Table 4.2).

Our goals were to generate a database of the sediment microbiome and, based on this, to develop a molecular bioindicator. Since elucidating a steady trend from our initial monitoring efforts, we selected several additional sites that will represent “test sites” outside of the Sdot Yam transect: Michmoret (38 m) and Ashdod (70 m). We also selected "disturbed sites": Herzliya marina (3 m) and the Hadera power

plant (6 m, Fig. 4.1) and characterized their sediment microbial communities. Importantly, those test sites selected for comparison (Ashdod and Michmoret) were consistent with trends observed for Sdot Yam across kingdoms, particularly regarding bottom depth (Fig. 4.2). Moreover, the sediment microbiota of Herzliya marina, a highly disturbed site, was markedly distinct in its composition across all kingdoms, which may indicate that the particular conditions in this site had more influence on the microbial community than the other parameters (bottom depths, seasons, etc.).

At the Hadera power plant site, where sampling was a considerable distance from the origin of the disturbance (a power plant outlet), the impact was better resolved in bacteria when compared to the two other kingdoms. The relative abundance of the most dominant bacterial taxa notably varied across bottom depth and core depth (Fig. 4.3a). In addition, we examined our ability to identify biomarker taxa, i.e., taxa that consistently explain the differences between communities, by analyzing sediment bacterial communities. Indeed, bacterial biomarkers were identified for each bottom depth (Fig. 4.3b) and season (Fig. 4.3c).

These results demonstrate that: 1) sediment microbiota present a complex and dynamic system and are robust indicators, making them an ideal model system for studying intra- and inter-kingdom interactions, 2) sediment microbiota may serve as a highly effective monitoring tool for environmental health, and 3) there remains a high potential to discover biomarkers for environmental changes and/or disturbances.

Figure 4.3: Composition of sediment bacterial communities. ***A****) Relative abundance distribution of dominant bacterial taxa. Amplicon sequence variants (ASVs) were binned to the genera level. Genera with prevalence of > 5% of samples and relative abundance of > 3% in at least in one sample were considered dominant and are presented here. Relative abundances were scaled by taxon and presented as a heatmap. Biomarker species of bottom depth (****B****) and season (C) are presented here. LefSE analysis*

presents the significantly discriminant taxa (P < 0.05, LDA effect size > 2). Classes represented by more than one genus are noted by color: Alphaproteobacteria, Gammaproteobacteria, Bacteroidia, Desulfobacteria and Planctomycetes.

5. Zooplankton community

Scientific supervisor: Dr Eli Shemesh

Table 5.1: Summary of zooplankton study, 2019-2021

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Plankton collected using a Bongo plankton net, 200 and 60 µm mesh.	Characterization of the plankton community, trophic position and carbon isotopic ratio data.	None	One season plankton identification and characterization of trophic position and carbon isotopic ratio data.	None

The Mediterranean Sea is an oligotrophic semi-enclosed basin whose marine life is threatened by the opening of the Suez Canal, the construction of the Aswan Dam, climate change, and additional anthropogenic factors. The opening of the Suez Canal caused the migration of marine species across the Suez Canal (Lessepsian migration), mainly from the Red Sea to the Mediterranean Sea.

Zooplankton are key organisms in the pelagic food web that shape the pelagic ecosystem. They transfer the organic compounds produced by phytoplankton and export them up the food web, and constitute a primary food source for many carnivores (Bouley and Kimmerer 2006).

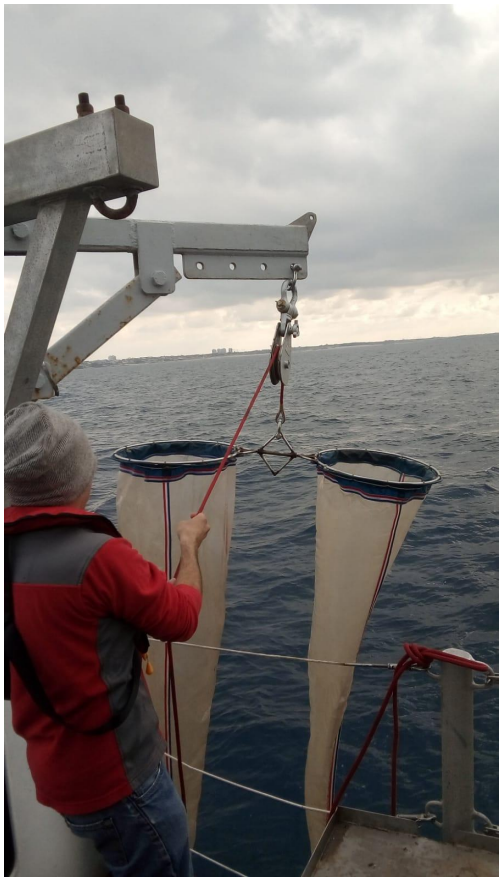
Marine zooplankton are rapid-responders to environmental variation, and are associated with regime shifts and climate change. Their shift in the ecosystem may cause significant and potentially accelerating losses in species diversity (Beaugrand et al., 2010; Mollmann and Diekmann, 2012). Most living organisms in the sea begin their life as plankton. Indeed, most will not reach the adult stage and instead serve as food for other marine animals. However, they are still indicative of the potential recruitment within our region. Our goal is to better understand the recruitment potential of marine

organisms, which can help us better understand the health of our environment and the resilience of different species.

Methods

Sample collection:

The plankton was collected on July 17, 2019, from three bottom depths near Sdot Yam: 25 m, 45 m and 100 m using a 50 cm diameter Bongo Plankton net (mesh sizes 60 and 200 µm). The bongo net was towed at three to five m depth in all locations.



Bongo net deployed off the Mediterranean Explorer research vessel

Molecular analysis:

Total genomic DNA of the plankton was extracted using the Wizard SV Genomic DNA Purification System kit (Promega). Partial sequences of the mitochondrial cytochrome c oxidase I (COI) and 18s (rRNA) genes were amplified using the Next Generation tagged primers mlCOIintF/jgHCO as used in Mora et al. (2011) and 18S 1389F\1510R primer by Alberti (2017), respectively. Using Next Generation Sequencing of Cytochrome Oxidase I (COI) and 18s (rRNA) markers, we identified the collected plankton.

Compound-specific stable isotope analysis:

Approximately 5 mg of lyophilized plankton sample was acid hydrolyzed in 1 ml of 6 nmol HCl at 150 $^{\circ C}$ for 75 min (Cowie & Hedges, 1992) under a nitrogen atmosphere inside a 4 ml glass vial with PTFE cap. Samples were cooled to room temperature, then HCl was evaporated under a gentle stream of nitrogen. Samples were neutralized twice with 1 ml ultra-pure water and under evaporation with a gentle stream of nitrogen. We used an EZ:faast amino acid analysis kit with a slight modification, replacing reagent 6 with dichloromethane as the solvent. For carbon analysis, we injected 1.5 µl in split mode (1:15) at 250 ^{0}C; for nitrogen, we injected 2 µl in split mode (1:5) at 250 ^{0}C. Helium was used as a carrier gas at a constant flow of 1.5 ml/min. The amino acids were

separated on a Zebron ZB-50 column (Phenomenex) in Thermo Scientific Trace 1300 GC. Gas chromatography (GC) condition was set to optimized peak separation for the desired amino acids as follows: an initial temperature of 110 ^{0}C was increased to 240 ^{0}C at a rate of 8 ^{0}C per minute. Then, the temperature was increased to 320 ^{0}C at a rate of 20 ^{0}C per min and held for 2.5 minutes. The separated amino acids were split on MicroChannel Device into two directional flows, one toward Thermo Scientific ISQ quadruple for amino acid identification and the second toward Thermo Scientific Delta V advantage for C and N isotope analysis. The ISQ condition was set to transfer line 310 ^{0}C, ion source 240 ^{0}C and scan range from 43 to 450 mass range. To define the isotopic ratio of carbon and nitrogen, the separated amino acids were combusted in a Thermo Scientific GC isolink II at 1000 ^{0}C for CO_2 and N_2. Before entering Delta V for the N_2 analysis of the sample, it went through a liquid nitrogen cold trap to freeze down all other gases. From each sample, duplicates were injected for carbon and triplicates for nitrogen.

Results

The results presented here cover the first plankton sampling campaign on the 17th of July, 2019. The sampling, molecular and isotopic methods were well executed, and thus we will use them in future plankton sampling. Since these are baseline results and we don't have data to compare them with, we present only the sequencing results of species collected, the carbon isotopic results, and the trophic position of the collected plankton at the different bottom depths. The two gene markers (COI and 18s) produced different results due to different affinity of the primers.

The COI relative abundance results show that the planktonic community from the July 17th sampling was mostly composed of crustaceans, followed by mollusca, fish and hydrozoan taxa (Figure 5.1 and Table 5.2). The 18s ribosomal RNA analysis was dominated by crustaceans and, to a lesser degree, marine worms (Figure 5.2 and Table 5.3). The species-specific data is presented in Plots 5.3- 5.8 and inTables 5.4 - 5.9.

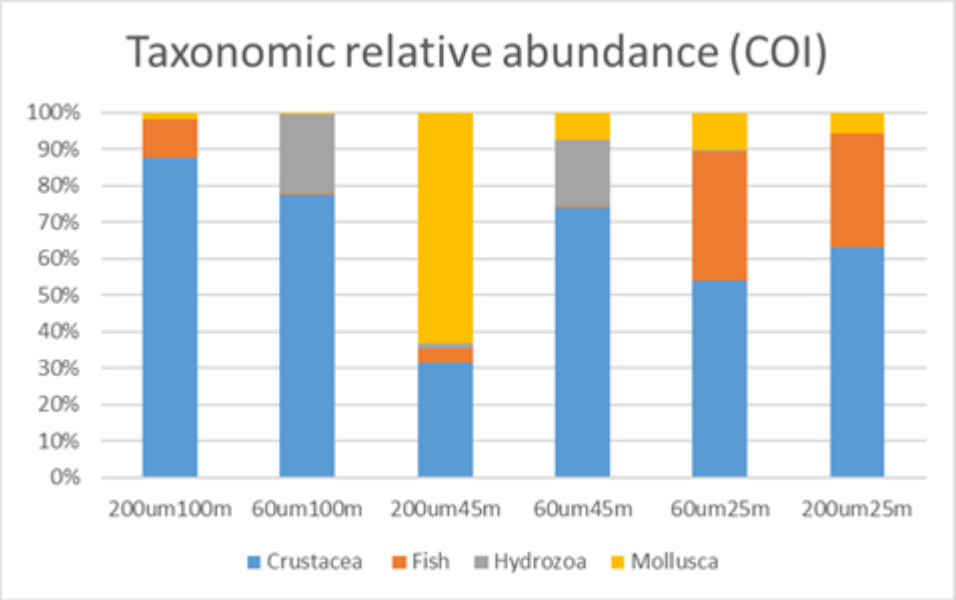


Figure 5.1 Relative abundance of the Operational Taxonomic Unit (OTU) from the partial Cytochrome Oxidase I (COI) gene of the zooplankton samples collected with 200 µm (200um) and 60 µm (60 um) meshes at 100 m, 45m and 25 m water depth stations.

Table 5.2 OTU numbers for each taxa within the study, oxidase I (COI) gene of the zooplankton samples collected with 200 µm and 60 µm meshes at 100 m, 45m and 25 m water depth stations.

Taxa	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Crustacea	2927	266	1021	1436	2273	2402
Fish	347	1	119	3	1494	1193
Hydrozoa	2	75	45	358	10	3
Mollusca	62	2	2042	144	441	223

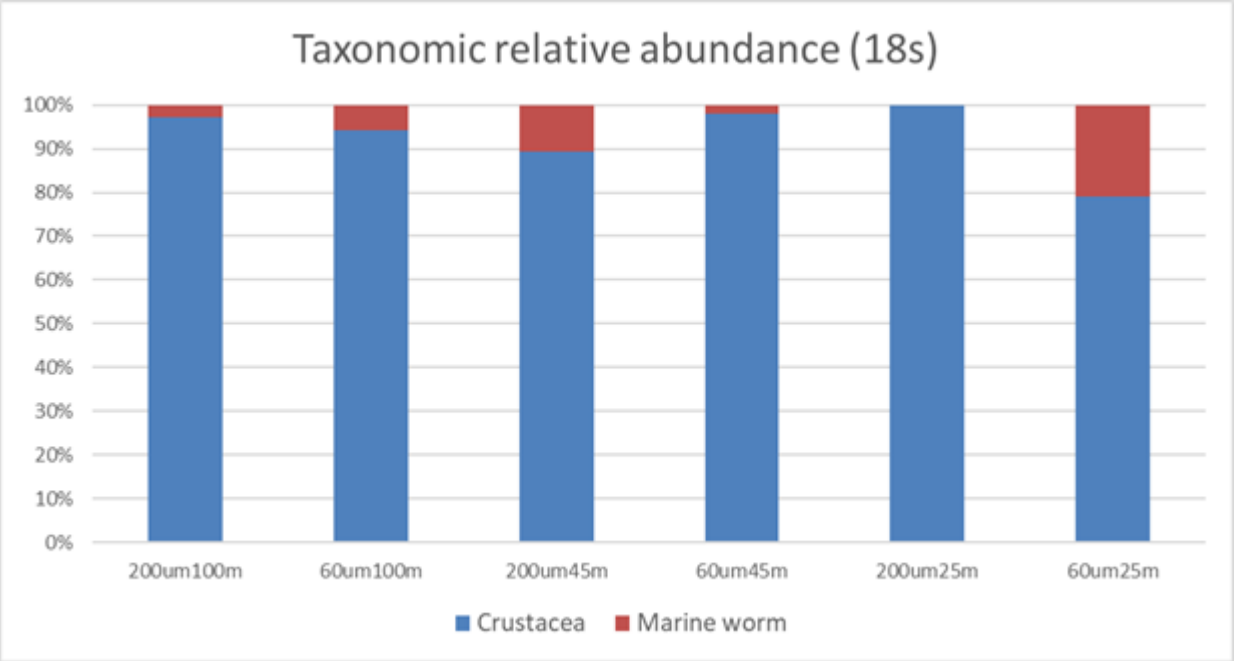


Figure 5.2. Relative abundance of Operational Taxonomic Unit (OTU) of partial 18s ribosomal RNA (rRNA) of the zooplankton samples collected with 200 µm and 60 µm mesh nets at the 100 m, 45m and 25 m water depth stations.

Table 5.3 Operational Taxonomic Unit (OTU) numbers of partial 18s ribosomal RNA (rRNA) of the zooplankton samples collected with 200 µm and 60 µm meshes at the 100 m, 45m and 25 m water depth stations.

Taxa	200µm 100m	60µm 100m	200µm 45m	60µm 45m	200µm 25m	60µm 25m
Crustacea	703	2964	503	2975	72	1031
Marine worms	20	186	60	64	0	273

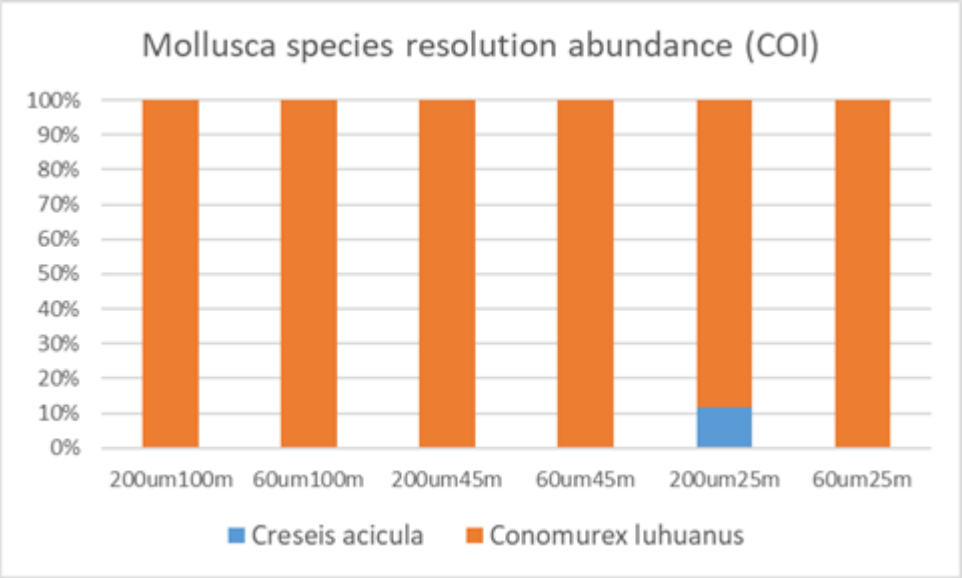


Figure 5.3 Operational taxonomic unit (OTU) relative abundance of Cytochrome Oxidase I (COI) gene of mollusca in the zooplankton samples collected with 200 µm and 60 µm mesh nets at the 100m, 45m and 25 m water depth stations.

Table 5.4 (OTU)numbers of Cytochrome Oxidase I (COI) gene of mollusca in the zooplankton samples collected with 200 µm and 60 µm meshes at the 100 m, 45m and 25 m water depth stations.

	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Creseis acicula	0	0	0	0	26	1
Conomurex luhuanus	62	3	2042	144	197	440

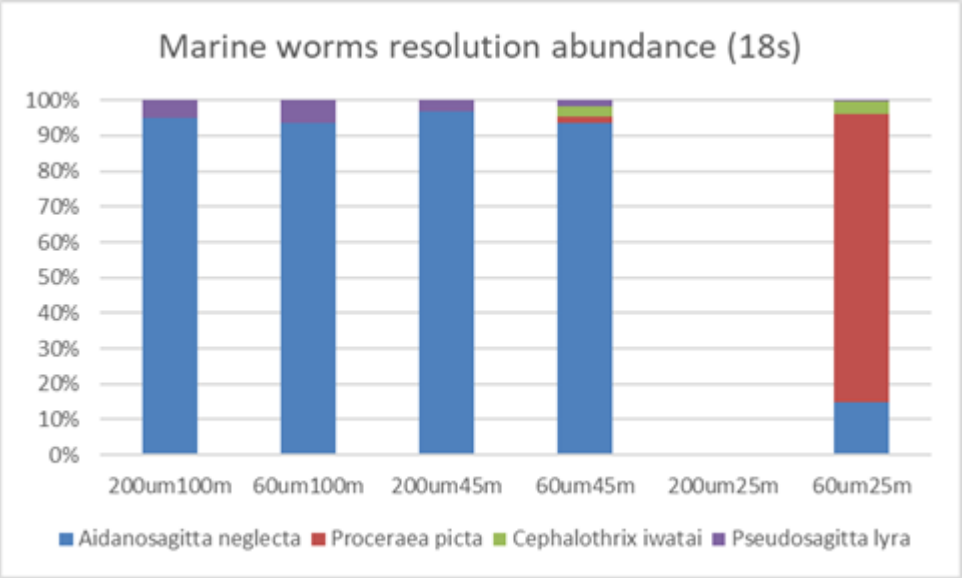


Figure 5.4. Operational taxonomic unit (OTU) relative abundance of partial 18s ribosomal RNA (rRNA) of mollusca in the zooplankton samples collected with 200 µmand 60 µm mesh nets at the 100 m, 45m and 25 m water depth stations.

Table 5.5 Operational taxonomic unit (OTU) numbers of partial 18s ribosomal RNA (rRNA) of mollusca in the zooplankton samples collected with 200 µm and 60 µm meshes at the 100 m, 45m and 25 m water depth stations.

	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Aidanosagitta neglecta	19	174	58	60	0	40
Proceraea picta	0	0	0	1	0	222
Cephalothrix iwatai	0	0	0	2	0	10
Pseudosagitta lyra	1	12	2	1	0	1

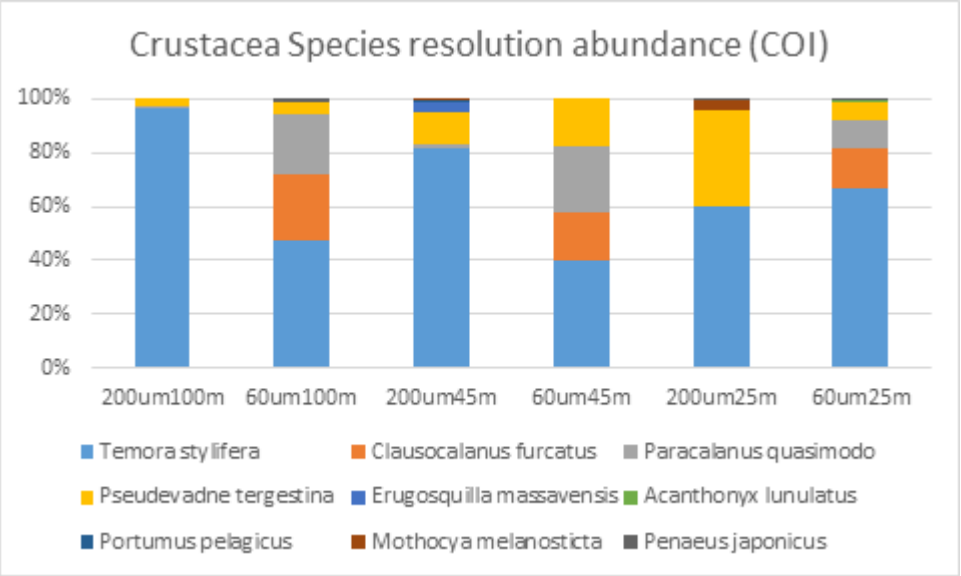


Figure 5.5. Operational taxonomic unit (OTU) relative abundance of Cytochrome Oxidase I (COI) gene of crustaceans in the zooplankton samples collected with 200 µm and 60 µm mesh netsat the 100 m, 45m and 25 m water depth stations.

Table 5.6 Operational taxonomic unit (OTU) numbers of Cytochrome Oxidase I (COI) gene of crustaceans in the samples collected with 200 µm and 60 µm meshes at the 100 m, 45m and 25 m water depth stations.

	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Temora stylifera	2826	155	834	557	1446	1514
Clausocalanus furcatus	6	80	3	250	3	339
Paracalanus quasimodo	21	75	9	334	2	238
Pseudevadne tergestina	73	15	126	246	844	147
Erugosquilla massavensis	0	0	36	0	0	0
Acanthonyx lunulatus	0	0	0	0	3	27
Portumus pelagicus	0	0	11	0	0	0
Mothocya melanosticta	0	0	2	0	96	0

Penaeus japonicus	1	3	0	0	8	2

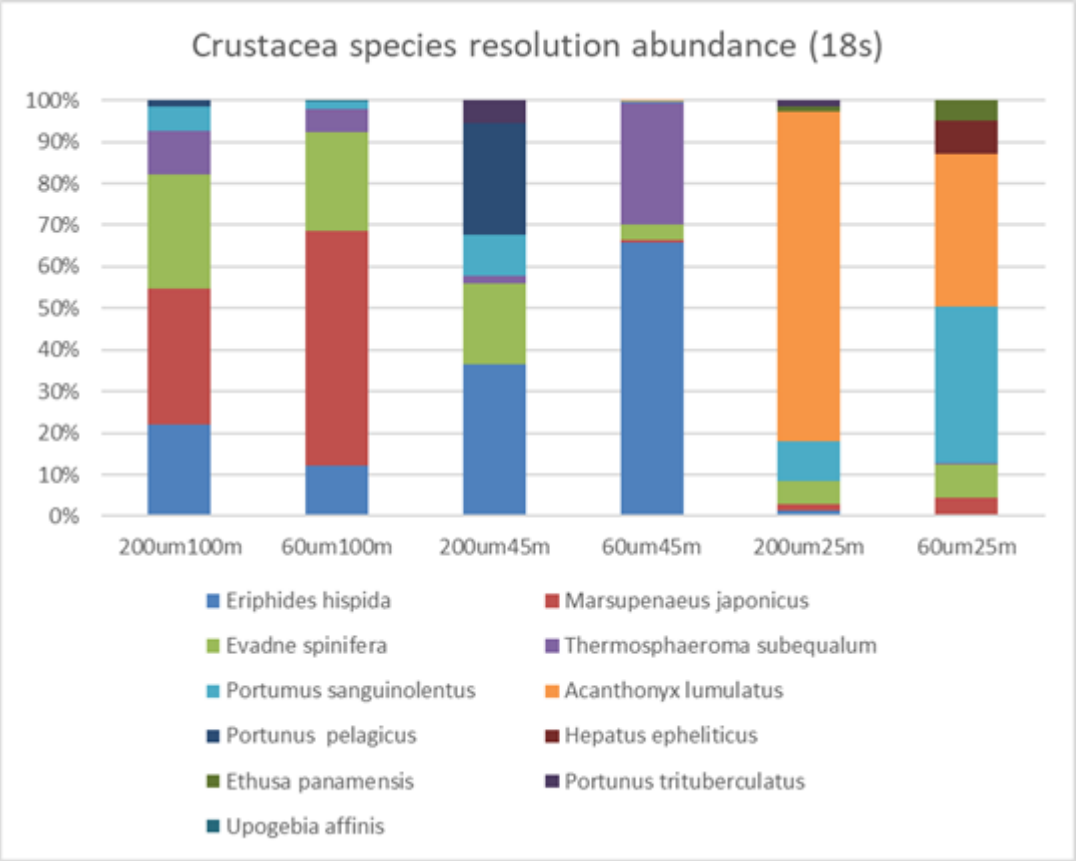


Figure 5.6. Operational taxonomic unit (OTU) relative abundance of partial 18s ribosomal RNA (rRNA) of crustaceans in the zooplankton samples collected with 200 µm and 60 µm mesh netsat the 100 m, 45m and 25 m water depth stations.

Table 5.7 Operational taxonomic unit (OTU) numbers of partial 18s ribosomal RNA (rRNA) of crustaceans in the samples collected with 200 µm and 60 µm meshes at the 100 m, 45m and 25 m water depth stations.

	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Eriphides hispida	154	365	183	1957	1	0
Marsupenaeus japonicus	231	1670	0	21	1	46
Evadne spinifera	193	705	98	106	4	83
Thermosphaeroma subequalum	73	159	10	871	0	2
Portumus sanguinolentus	41	57	50	13	7	389
Acanthonyx lumulatus	0	0	0	6	57	377
Portunus pelagicus	11	0	134	0	0	0
Hepatus epheliticus	0	0	0	0	0	84
Ethusa panamensis	0	0	0	1	1	50
Portunus trituberculatus	0	0	28	0	1	0
Upogebia affinis	0	8	0	0	0	0

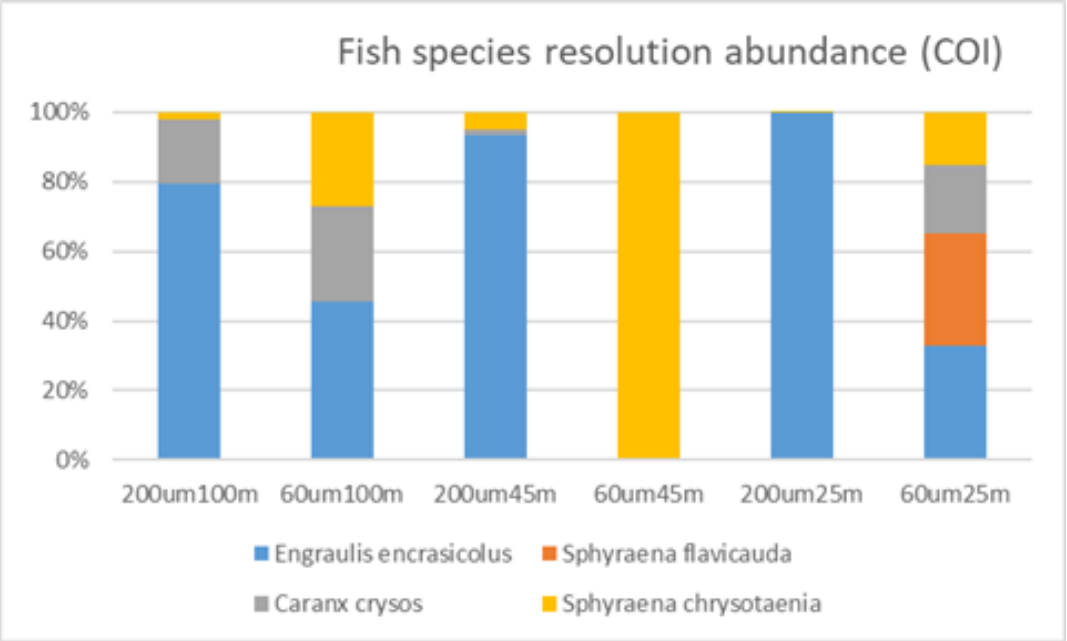


Figure 5.7. Operational taxonomic unit (OTU) relative abundance of Cytochrome Oxidase I (COI) gene of fish in the zooplankton samples collected with 200 µm and 60 µm mesh netsat the 100 m, 45m and 25 m water depth stations.

Table 5.8 Operational taxonomic unit (OTU) numbers of Cytochrome Oxidase I (COI) gene of fish in the samples collected with 200 µm and 60 µm mesh nets at the 100 m, 45m and 25 m water depth stations.

	200 µm 100 m	60 µm 100 m	200 µm 45 m	60 µm 45 m	200 µm 25 m	60 µm 25 m
Engraulis encrasicolus	275	5	111	0	1190	494
Sphyraena flavicauda	0	0	0	0	0	480
Caranx crysos	65	3	2	0	0	292
Sphyraena chrysotaenia	7	3	6	3	3	228

Isotopic Results

The trophic position was calculated from the equation TPGlu/Phe=((δ15NGlu–δ15NPhe–β)/TDFAA)+1 where β=-0.509 and TDFAA=4.46 (equation produced by Dr Stephane Martinez).

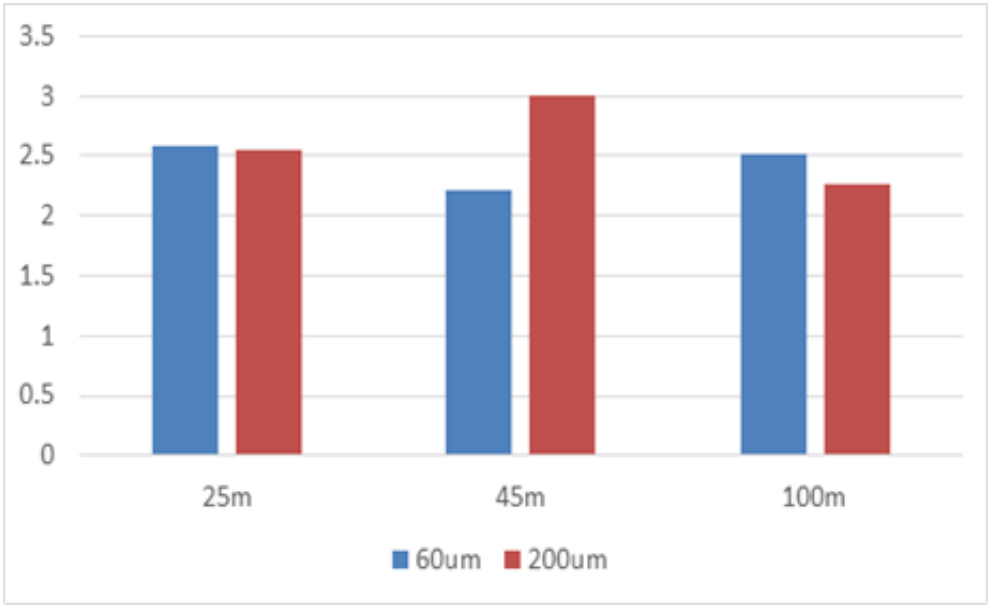


Figure 5.8: Trophic position (delta N) of the collected plankton from different depths and mesh sizes.

ANOSIM analysis of carbon isotope composition for five essential amino acids (Valine, Leucine, Isoleucine, Methionine, and Phenylalanine) in the planktonic community point to a significant difference between the 60 µm to the 200 µm net communities, without correlation to bottom depth.

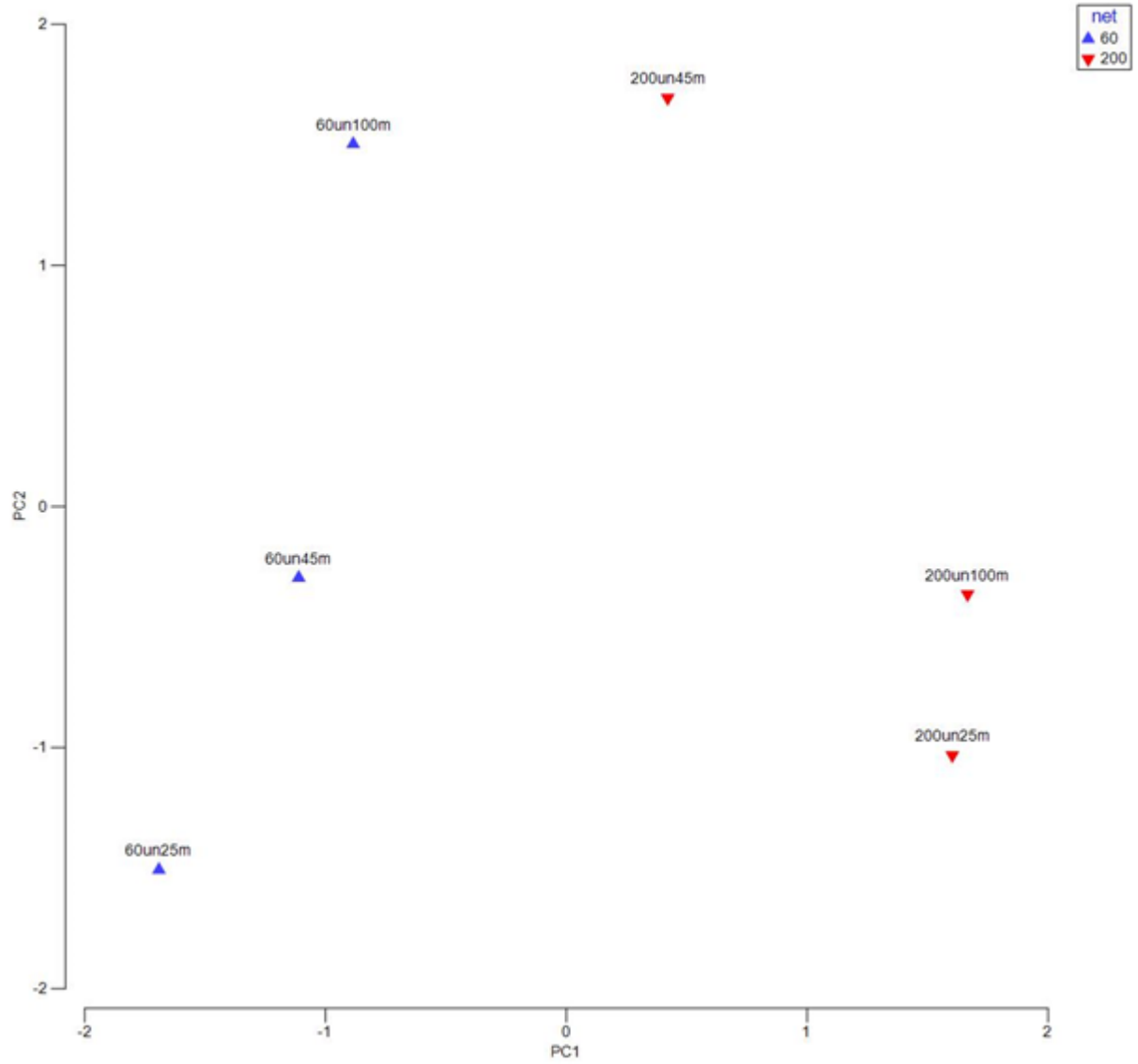


Figure 5.9: Carbon isotope PCA of five essential amino acids (Valine, Leucine, Isoleucine, Methionine, and Phenylalanine).

6. Biodiversity of the rocky reef

Leading scientist: DrStephane Martinez and Mr Hagai Nativ

Table 6.1: Summary of activities and biodiversity of the rocky reef (2014-2020)

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Rocky reef fish transect survey of species, abundance, and size distribution	Fish data collected from2015.	Three transects per site on the first season of sampling; Missing surveys at Ashdod and Ashkelon due to poor visibility	1. Marked seasonal differences in the number of fish at all sites (due to larger shoals *of Chromis chromis* and *Siganus rivulatus*).	
	Conducting four transects at each rocky reef site		2. A general increase in grouper numbers	
			3. The formation of invasive populations of *Torquigener flavimaculosu* and *Pterois miles*.	
			4. At Achziv reserve, small *Epinephelus marginatus* inhabit the shallower depths, while the fish size increases with depth	
Photo point sampling of coverage	Spring and Autumn surveys in all LTER rocky sites		1. The rocky reef is mostly covered in turf algae and an unidentified bryozoan colony	Algal and bryozoan specimens are currently in identification process
			2. A trend of decrease in algal cover and increase in bryozoan colonies and, to a lesser degree, sponge and ascidian taxa.	
			3.Deeper (45 m) sites are richer and more diverse than 10 m and 25 m sites	

The rocky reef habitat of Israel's coastline is rich and diverse. It functions as a stable habitat for algae, invertebrates, and fish. However, our knowledge of this important habitat is very limited and, until now, the research scope has been limited. Since 2014, we have been collecting data on the reefs of Achziv, Nahariya and Sdot Yam at three different depth levels (10, 25 and 45 m). These depths are safely surveyed using closed circuit diving gear, with minimal interruption to the fish.

During 2017, we added the sites of Ashdod and Ashkelon, surveying at 30 m depth, In 2019, we added shallow depth sites (up to 4 m) in Ashkelon, Sdot Yam and Achziv via cuba diving. We collect coverage data on invertebrates and algae, and visually survey the fish. We intend to follow changes in space and time along our coastline and hope that this knowledge will encourage researchers to investigate this habitat. The data serves as a good baseline for further research and enhances our knowledge of the marine ecosystem and application towards a more effective policy in Israel. We began by photographing fish along a 25 m transect line. . Then, we photographed quadrats (25 cm by 25 cm) at 2 m intervals. Fish are identified, counted, and sizes are annotated on each transect.

Methods

The aim of the rocky substrate fish survey is to follow up on changes in abundance and community structure along the Israeli Mediterranean coast. For this purpose, the visual survey method of Jennings & Polunin (1995) was used despite the possibility of its inherent bias. This method is suitable for most of the more abundant rocky habitat species (Guidetti et al., 2014), and was chosen to avoid the destruction of capture or poisoning methods. During the second season, video became our registry tool instead of the pencil and board, followingBrokovich et al. (2 008). Videography enables watching previous surveys as well as consultation with experts for the identification of newly encountered species.

Closed circuit divers during a fish survey (left diver) and during algae and invertebrate survey (right), using the photo-quadrat method along a transect line.

The same qualified surveyor is responsible for all fish transects since 2015. For the fish census, four transects of 25 m' each were conducted at each site. Every transect is recorded on video and all fish counts are spoken out loud for the recorder. The surveyor records species, number of individuals, size, and distance from the transect line on the first pass. On the way back, the benthic species are counted by placing a photo-quadrat (25cm by 25 cm) every 2 m along the transect. The photos are taken with

Canon G1X with 2 external flashes (Ikelite DS51). All photos are uploaded to the Coralnet website for analysis to generate stratified random points (4 rows x 4 columns of cells, 1 point per cell (a total of 16 points per photo).

The photos are automatically annotated by a machine learning program which assesses substrate coverage by functional groups and taxa, when the confidence level of machine recognition is higher than 90%. The remaining points are analysed and manually entered. The source photos and point data are available to the public at https://coralnet.ucsd.edu/source/307/. Data and basic analysis is also provided at: http://med-lter.haifa.ac.il/index.php/data-base/marine-invertebrates.

Since the photo quadrats do not sample all the species, we use a complementary method known as 'Time per Effort - Invertebrates'. Using this method, the diver swims freely for 10 minutes and lists every species seen (results not shown here). Unidentified species are collected for identification.

6.a Fish community results

Epinephelus marginatus, *a predator of the rocky reef*

Our long-term study has enabled us so far to identify a few phenomena in the rocky reefs, some of which we will describe in this chapter.

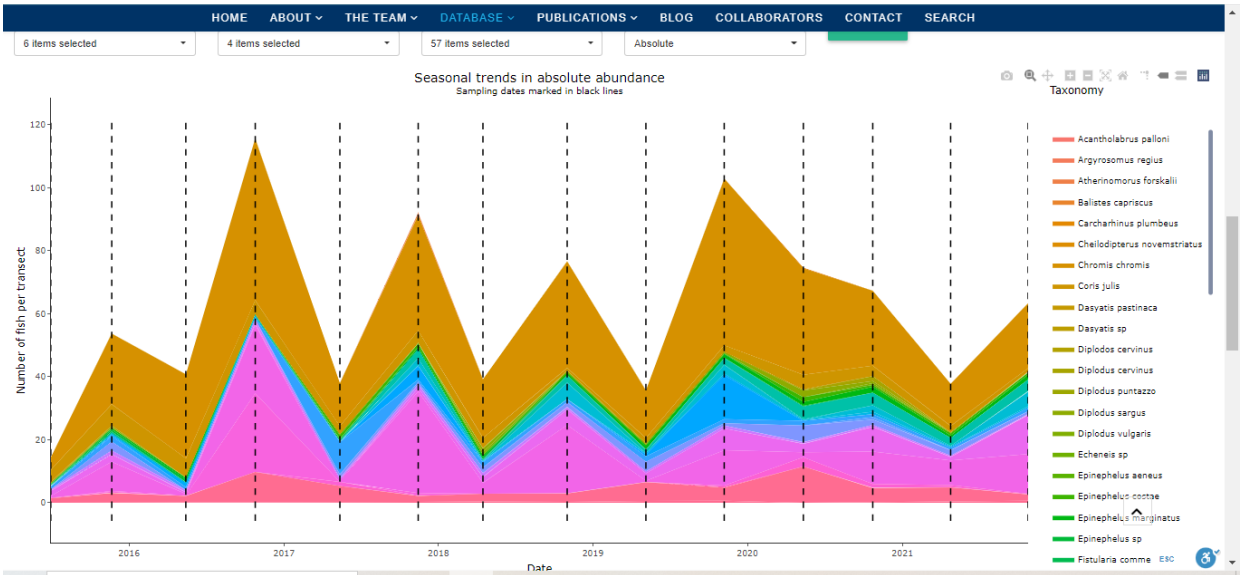


Figure 6.1 Number of fish at all sites and depths per sampling date from 2015-2021. Each surveying date is demarcated as a spotted vertical line. Sampling effort was lower in the first season; all the peaks occur during the autumn. A better view with the complete legend (and more interactive options) are available at: https://med-lter.haifa.ac.il/index.php/en/data-base/rocky-reefs/fish-survey

There are marked seasonal differences in the number of fish (Figure 6.1) across year and season. Shoals of *Chromis chromis* and *Siganus rivulatus* during autumn contribute most to these differences.

Throughout our study, we have witnessed a gradual increase in the number of grouper (*Epinephelus marginatus, Mycteroperca rubra*, *Epinephelus costae, Epinephelus aeneus)* fish numbers (Figure 6.2); In the Achziv 45 m depth at the marine protected reserve, we did not see a change .

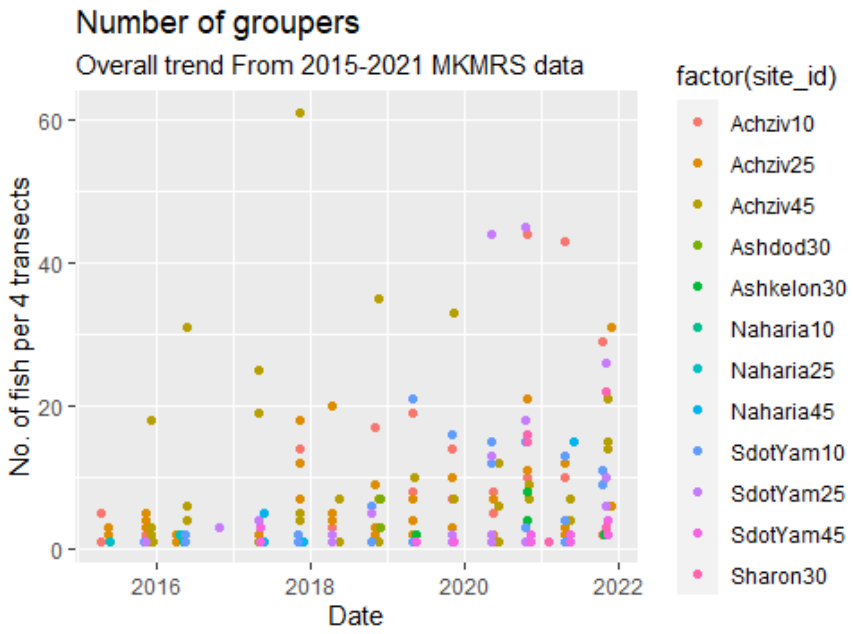


Figure 6.2: Number of grouper fish: Epinephelus marginatus, Mycteroperca rubra, Epinephelus costae, and Epinephelus aeneus in the rocky reef study sites.

Another long-term phenomenon we observed is the formation of invasive populations of *Torquigener flavimaculosus* and *Pterois miles* beginning in 2016 *(*Figures 6.3, and 6.4).

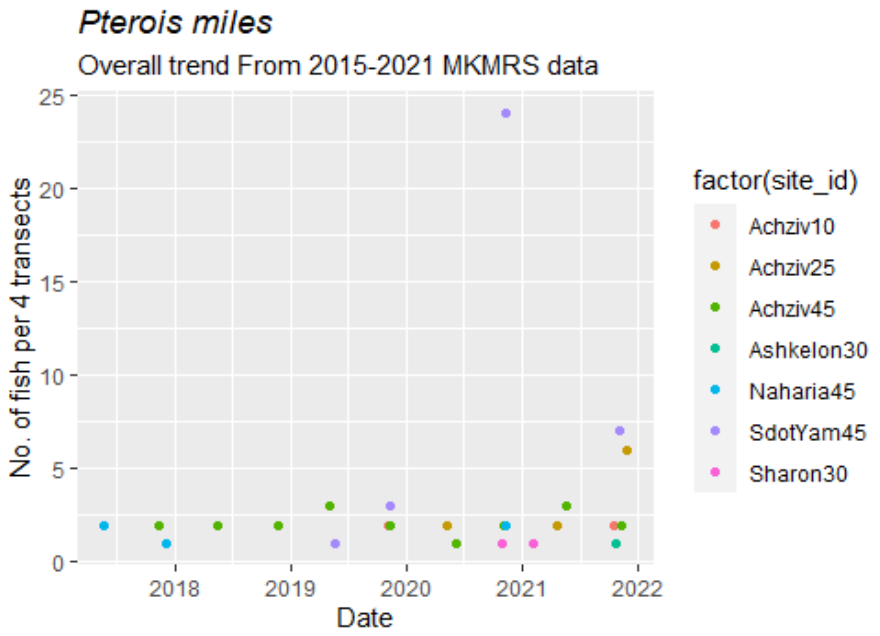


Figure 6.3: The formation of an invasive population of Pterois miles*, at the study sites.*

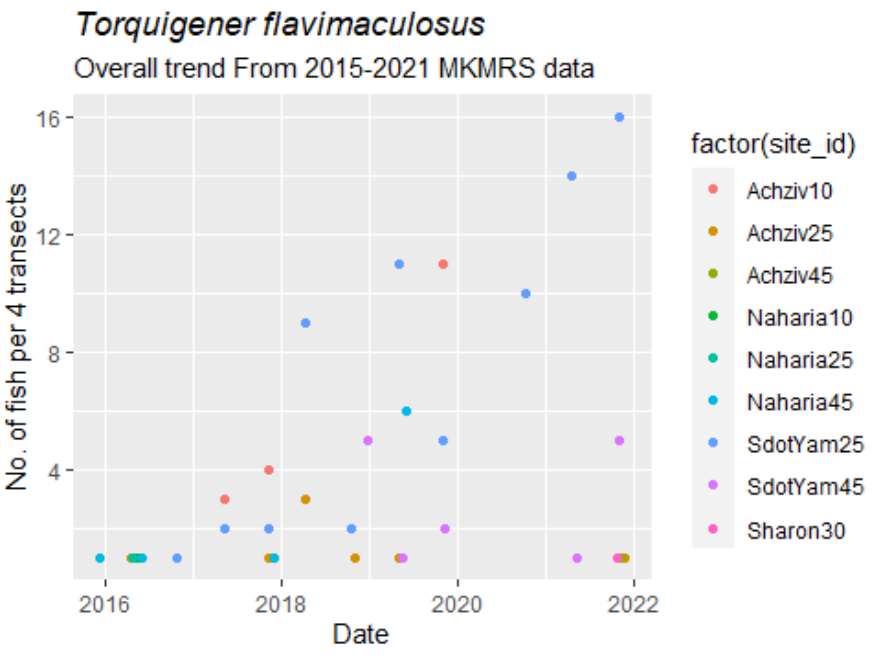


Figure 6.4: The formation of an invasive population of Torquigener flavimaculosus*, at the study sites.*

Torquigener flavimaculosus - a very successful Lessepsian invader

The study within the Achziv reserve has shown us that the shallower depth sites are a nursery for the predator fish *Epinephelus marginatus,* whereas the larger sized fish reside in the deeper parts of the reserve (Figure 6.5).

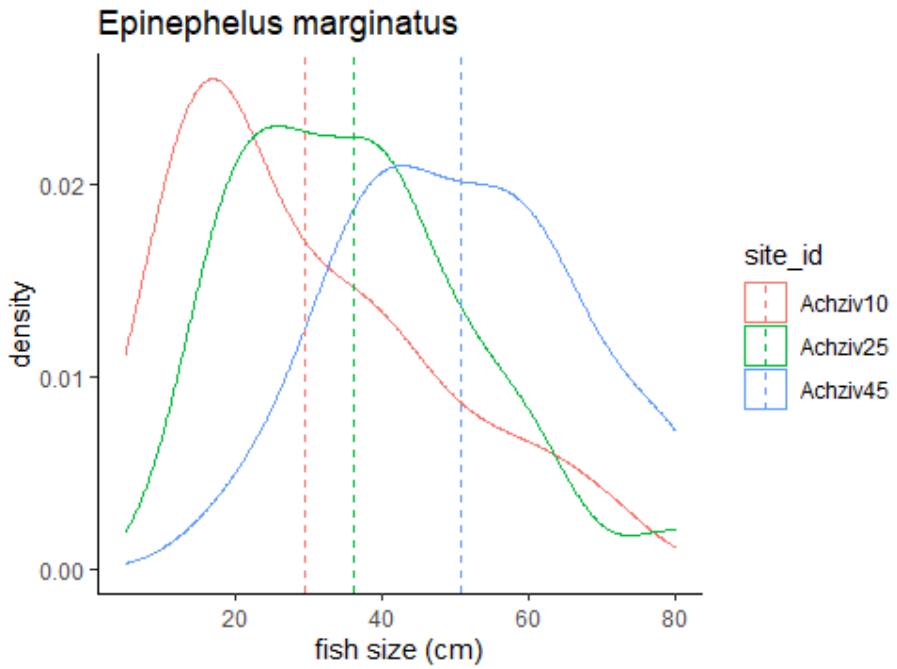


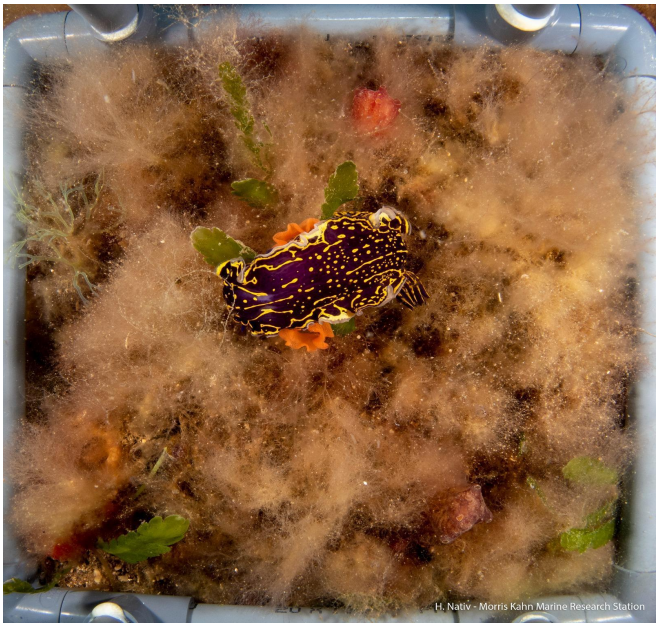
Figure 6.5 Size distribution of predator fish Epinephelus marginatus in 10, 25 and 45 m depth rocky sites at the Achziv reserve 2015-2021. Average fish size is represented in a dotted line.

6.b Invertebrate and algal community

Species identification: Dr Sigal Shefer and Dr Noa Shenkar

Methods

All photos are uploaded to the Coralnet website to undergo point sampling analysis.



A photo-quadrat from the Hof Hasharon 30 m, May 2022, dominated by an algae canopy of Dictyota sp., Lophocladia, and Caulerpa mexicana.

The photos are annotated automatically by a machine learning program which assesses substrate coverage by functional groups and taxa, and the confidence level of machine recognition is higher than 90%. The remaining points are analysed and manually entered. The source photos and point data is open to the public at https://coralnet.ucsd.edu/source/307/. Data and basic analysis is also provided at: http://med-lter.haifa.ac.il/index.php/data-base/marine-inverterates.

Results

A photo-quadrat from Ashdod 30 m, May 2022, dominated by an unidentified bryozoan

Since 2015 and until the end of 2021, we have collected over 6,000 images of benthos to document the algal and invertebrate coverage and community over the rocky reef.

The rocky substrate is dominantly covered by turf algae, followed by an unidentified branching bryozoan, tenfold as much as any

other substrate.

We have been witnessing a general decrease in algae, an increase in cover of unidentified branching bryozoan colonies (Fig. 6.6), and an increase in sponge (Figure 6.7) and ascidian cover.

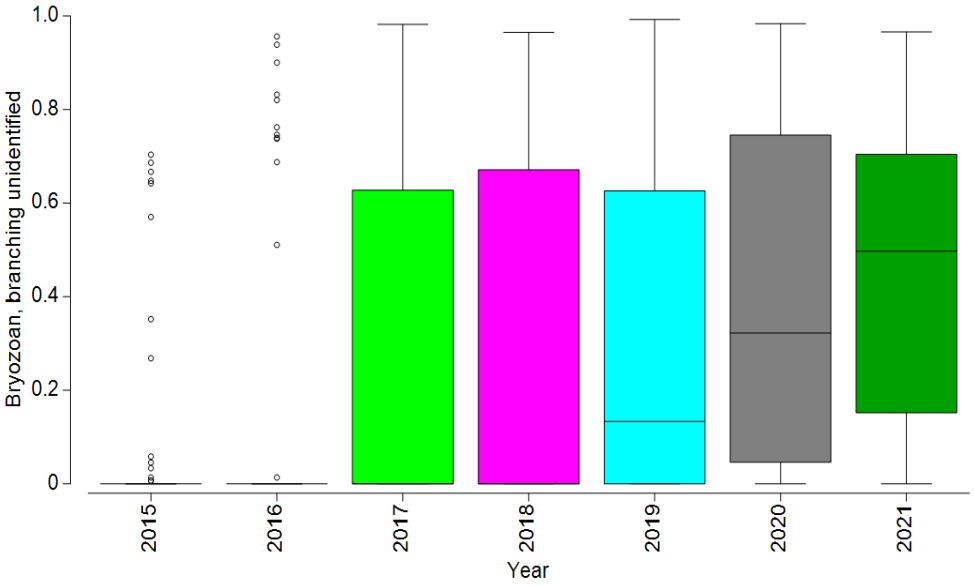


Figure 6.6 A box plot of relative abundance (scale of 0-1) of an unidentified branching bryozoan, from coverage assessment by photo quadrat point sampling, from all long-term research sites. The box plot represents the 1st and 3rd quartiles, and the band inside the box is the median. The ends of the whiskers represent the minimum and maximum of the data. Outliers are depicted as circles.

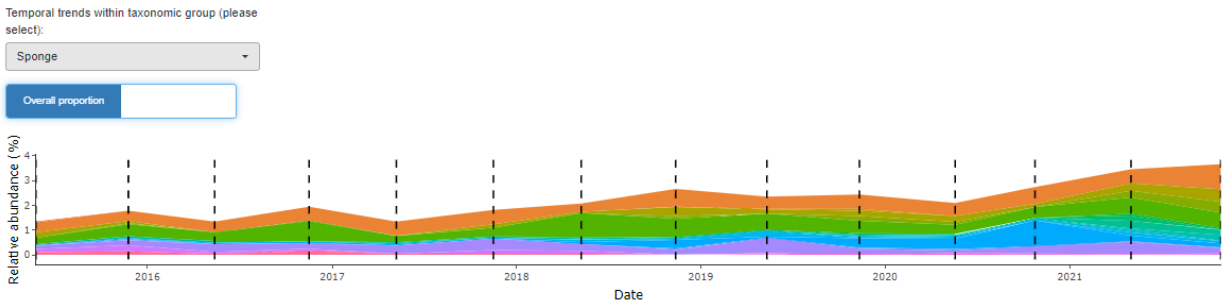


Figure 6.7 Sponge increase in relative abundance with time. This data can be further explored at: https://med-lter.haifa.ac.il/index.php/en/data-base/rocky-reefs/marine-invertebrates

The rocky reef is by no means uniform in terms of diversity. The deep (45 m) sites are more rich and diverse than the shallower reefs (Figures 6.8-6.9). These are preliminary and robust insights from our database, which have yielded questions, however, that is outside the scope of this report.

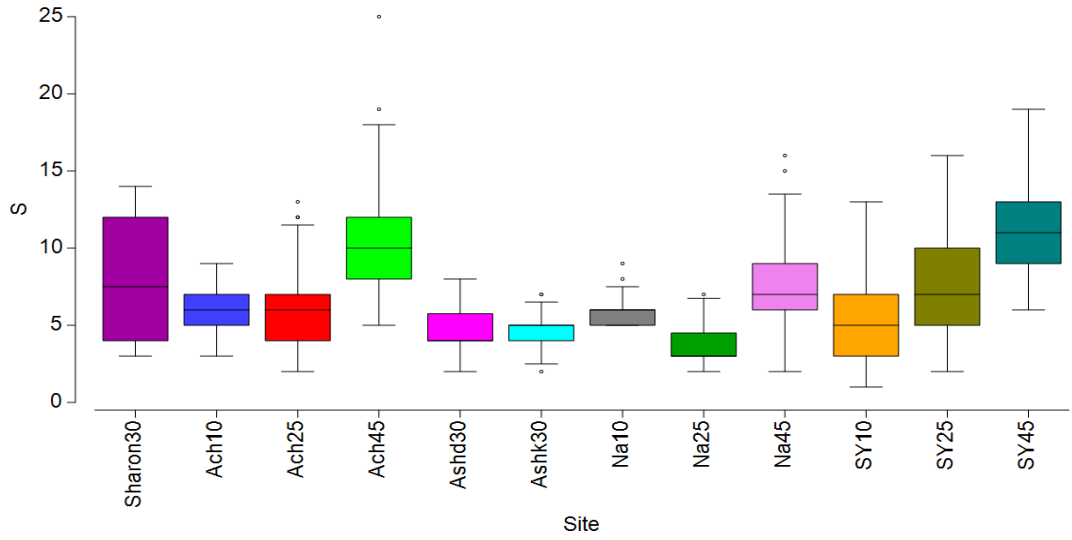


Figure 6.8 Box plot of invertebrate and algal species richness (S) in the different rocky reef sites, pooled year and season data. Ach- Achziv, Na- Nahariya, Ashd- Ashdod, Ashk- Ashkelon, SY- Sdot Yam. The depth of the sampling site (in m) is the suffix of the acronym.

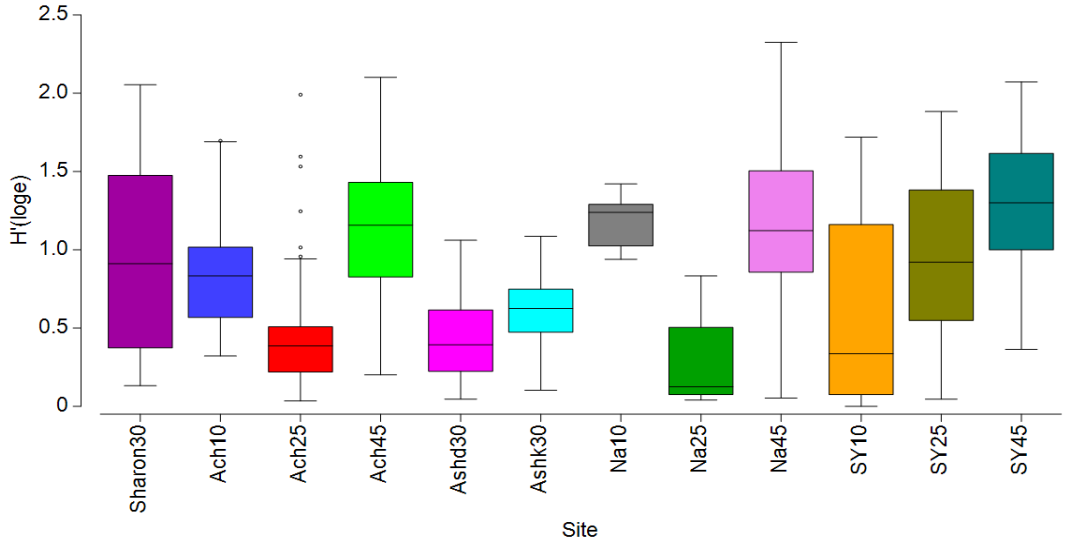


Figure 6.9 Box plot of Shannon-Wiener diversity index (H'log(e)) in the different rocky reef sites, pooled year and season data. Ach- Achziv, Na- Nahariya, Ashd- Ashdod, Ashk- Ashkelon, SY- Sdot Yam. The depth of the sampling site (in m) is the suffix.

7. Apex Predators

Leading scientist: Dr Aviad Scheinin

Dusky shark (*Carcharhinus obscurus*) in the outlet of the Orot Rabin Power Plant in Hadera, Israel

Table 7.1: Summary of activity for the Apex Predator lab, 2015-2021

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Shark survey, tagging and reproductive studies (*Carcharhinus obscurus* and *Carcharhinus plumbeus)* in Hadera.	Morphometrics; haematology, biochemistry and genetics; reproduction; passive acoustics satellite telemetry.	None - the satellite tags due to barnacle build-up in warm waters, or premature release as a result of fishing pressure; lack of ultrasound for recording of reproductive state.	67 sharks have been tagged. Acoustic data suggests site fidelity of both species to anthropogenic hotspots.	Consider the use of SPOT or camera tags over MiniPats (archival tags) to ensure successful reporting.
				Progress to understanding the reproductive cycle of the dusky sharks.
				Non-reliance on ultrasound for next season; renting EVO II portable ultrasound for autonomy.
	Abundance data, spatial and temporal use of the area.	Sea conditions were not suitable for sampling or surveying at times.	Characterized Ma'agan Michael as a nursery ground for the blackchin guitarfish.	Visual abundance surveys by drone.
				Long-term follow-up on Ma'agan Michael and Evtach sites.
Acoustic monitoring of coastal dolphin populations	Continuous presence/absence data in five locations: Nahariya, Dor, Michmoret fish farms, Ashdod fish farms and Ashkelon.	Finding a safe position for the F/C-Pod receivers is a major challenge.	Continuous presence/absence day and night data in the vicinity of the receivers (Approximately 1 mile).	Having a larger network of receivers will produce a better map of coastal dolphin's distribution.
			Continuous data on underwater background noise.	Try to differentiate between the two coastal dolphin species as detected by the F-Pod

Table 7.1: Summary of activity for the Apex Predators lab, 2015-2020- CONTINUED

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Bluefin tuna and pelagic megafauna	Habitat model for the Atlantic bluefin tuna in the eastern Mediterranean Sea and relative trends of abundance		Currently ongoing. Conclusions will be drawn at a later stage.	Conclusions will be drawn at a later stage.
Method development	Protocol design and standardization of aerial drone survey methods, underwater active acoustic survey methods	Gaps between the different schools working on this project (e.g., engineers and statisticians).	Automated image analysis and active hydro-acoustic detection technologies have been developed as well as analysis algorithms. These are currently being tailored for large-scale ecological surveys. Not detailed in this report.	Further improvement of the acoustic algorithm(s) and sea-trials for target separation and detection distance.
				Compare numeric models with controlled experiments in laboratory conditions.
				Determine the ideal coverage area to resolution ratio for accurate group size of marine mammals in drone footage.
Method development	e-DNA	Technical problems with water filtering and DNA extractions	Fish DNA was detected in a few locations. Analysis is performed at time of report's publication.	Working on improving the sampling protocol and sample preparations.
	Detection of marine vertebrates' presence			

The Apex Predator project deals with the development of sampling protocols and indicators for pelagic megafauna, with a focus on sharks and rays, bluefin tuna, and coastal dolphins, in a long-term ecological research framework. Emphasis is put on the function of these species as sentinels of the marine environment and their ability to inform about human perturbations. We employ photogrammetric measurements using drones and unmanned aerial vehicles (UAVs), active and passive-acoustic surveys, mark-recapture methods, underwater observations, and cutting-edge technologies for molecular and stable isotope analyses. Data and basic visualisation is provided also in the Marine Apex Predators database.

In addition to the long-term study, our lab is collaborating on several research projects. For example, we played a key role in the European Commission's 'Horizon 2020' programme. The project focused on remote sensing of fish by means of active hydro-acoustics, and enhancing mobile target detection based on swimming behaviour and size. The project was led by Dr Roee Diamant from the Department of Marine Technologies at the University of Haifa.

7.a Shark tagging and surveys in Hadera

In the winter months (November to May) since 2016, approximately 40–80 sharks (dusky shark, *Carcharhinus obscurus*, and sandbar shark, *Carcharhinus plumbeus;* Shamir et al., 2019) have been aggregating very close to the shore on Israel's Mediterranean coast, near the power plant in Hadera (32°27' N, 34°52' E). This phenomenon is unique to this region and provides an opportunity for the study of 'data deficient' and 'endangered' species, respectively, that are no longer observed in other parts of the Mediterranean Sea where they were once abundant. Furthermore, these two shark species from the Genus *Carcharhinus* form a winter mixed-species aggregation, which is also unique and interesting from a behavioural and ecological point of view.

Results

To date, 67 sharks have been captured and released at the foot of the coastal power plant in Hadera. Of these, 48 individuals belonged to the species *Carcharhinus obscurus* (34 females, 1 male) and 20 were of *Carcharhinus plumbeus* (all males except 1). These species are commonly referred to as dusky and sandbar sharks, and are listed by the

IUCN as "Data Deficient" and "Endangered" in the Eastern Mediterranean Sea, respectively. Forty-nine acoustic tags have been surgically implanted and ten satellite archival tags (MiniPats, Wildlife computers) were attached to individuals of both species. Tag-derived data are currently being analysed by the lab's students.

A total of 55,827 detections were recorded by moored acoustic receivers. The mean ± SD total length of tagged female dusky sharks was 293.97 ± 29.34 cm TL (with the unique male measuring 263 cm TL) and was 181.13 ± 8.29 cm TL for male sandbar sharks.

The mixed-sex community was composed of females mostly, and males seen during the last third of the season when the presence of sharks is relatively high (see Fig 7.1 below).

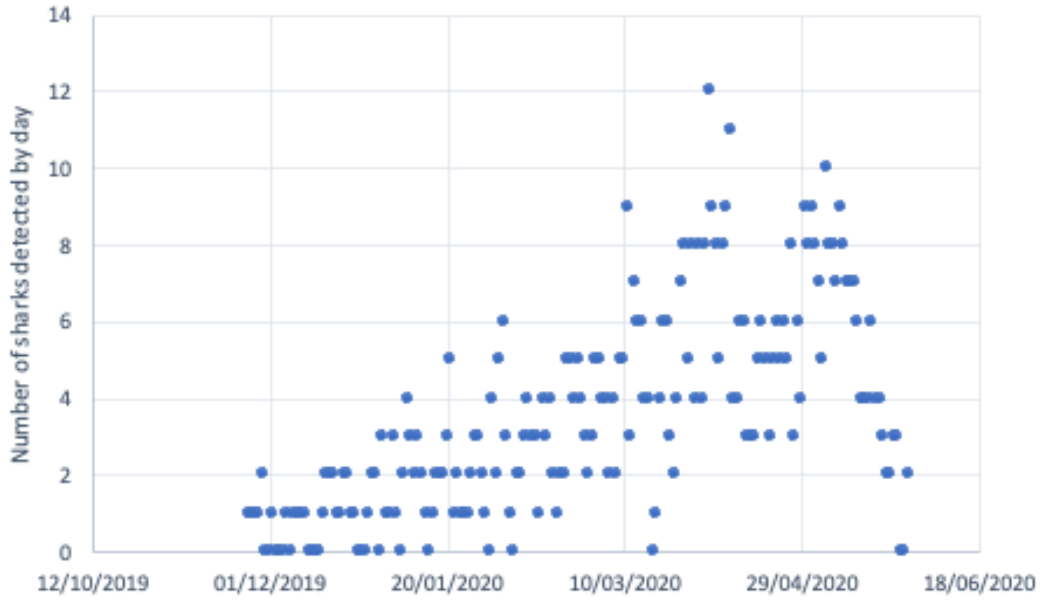


Figure 7.1 Number of sharks detected per day. Blue dots represent the number of sharks detected at least once on each day.

Preliminary results suggest year-round site fidelity to the power plant and an offshore fish farm in Ashdod, as well as the occupancy of other areas beyond the Mediterranean marine space of Israel. For example, tagged shark number 10 (ZLILA) left the Hadera aggregation on April 14, 2019, and travelled to Ashdod on April 23, 2019. After a short stay, she returned to Hadera and resided there until the end of May. We also noted that the male dusky shark (the only male tagged) left the area on April 30, 2019, at 01:24 am and was detected in Ashdod on the same day, at 18:01 pm.

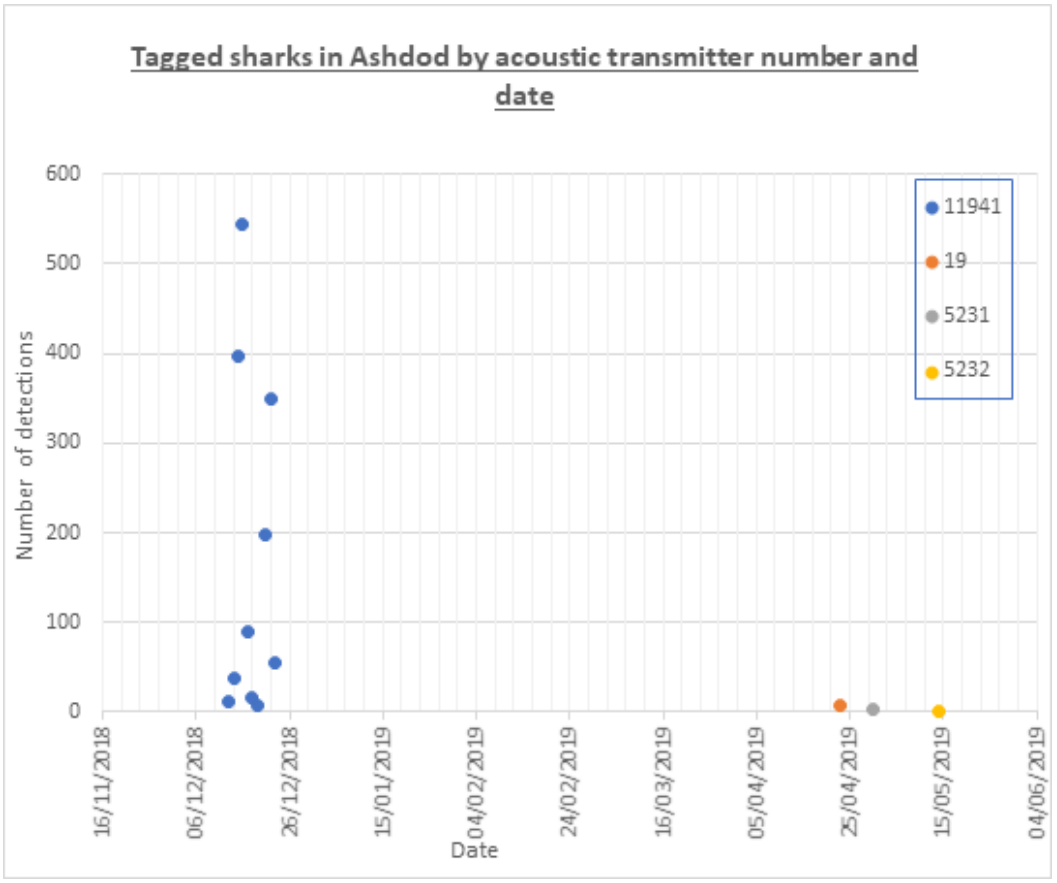


Figure 7.2. Number of detections per tagged shark (see legend), in Ashdod by date.

This data is available for download at:
https://med-lter.haifa.ac.il/index.php/en/data-base/marine-top-predators/shark-tagging

7.b Blackchin Guitarfish (Glaucostegus cemiculus) coastal survey

In the past few years, observations were made of young guitarfish along the shallow depths near Kibbutz Ma'agan Michael and along Evtach MPA on the Israeli Mediterranean coast. In this region, the young guitarfish exhibits uncommon behaviour, and can be found ascending the beach and staying outside of the water for short periods of time before returning to the sea. The guitarfish project started in 2017 and is focused on the endangered common guitarfish (*Rhinobatos rhinobatos*) and the critically endangered Blackchin guitarfish (*Glaucostegus cemiculus*), and found that Ma'agan Michael coastal area qualifies as a nursery ground for these species. This was done using capture-sampling-release protocols, and weekly visual surveys during the hypothesised pupping season (June-December) and once a month during the rest of the year. This work is continued in Poleg and Evtach. The observer counts are documented on a designated mobile app while the observer is walking along the shore at a consistent

pace.

Neonate guitarfish (A) in the intertidal zone

Results

To date, 125 visual abundance surveys have been conducted. In our study, a total of 549 guitarfish have been captured, measured, and released over 37 field days and with 215 net deployments. Of these 549 specimens, 327 have been tagged with PIT tags and 9 recaptured. Genetic verification has been performed on 41 specimens; 34 from Ma'agan Michael, six specimens from Evtach, and a single specimen of a common guitarfish for comparison. Genetic analysis showed that 40 of the specimens captured were positively identified as IUCN critically endangered blackchin guitarfish, and the single sample of the common guitarfish was positively identified as a common guitarfish. The guitarfish captured ranged from 23 cm to 81.3 cm in total length (mean: 34.9 ± 6.3) and weighted between 45 grams to 1810 grams (mean 144.9±145.7). Based on umbilical scar presence in all individuals at the beginning of the pupping season, size at birth was estimated to be 31.9 cm. Tagged, recaptured individuals showed a growth rate of approximately 1.9 cm per month. The data gathered thus far characterizes the Ma'agan Michael area as a nursery ground for neonate blackchin guitarfish. in addition, Evtach MPA was visually surveyed since 2018 until this day on a monthly basis and results from this site correlate to the result from Ma'agan Michael in the temporal distribution, suggesting that Evtach also functions as a nursery ground for the blackchin guitarfish. Other sites that were examined during this study are Krayot area and Poleg marine MPA, in both sites there was little to no presence of guitarfish.

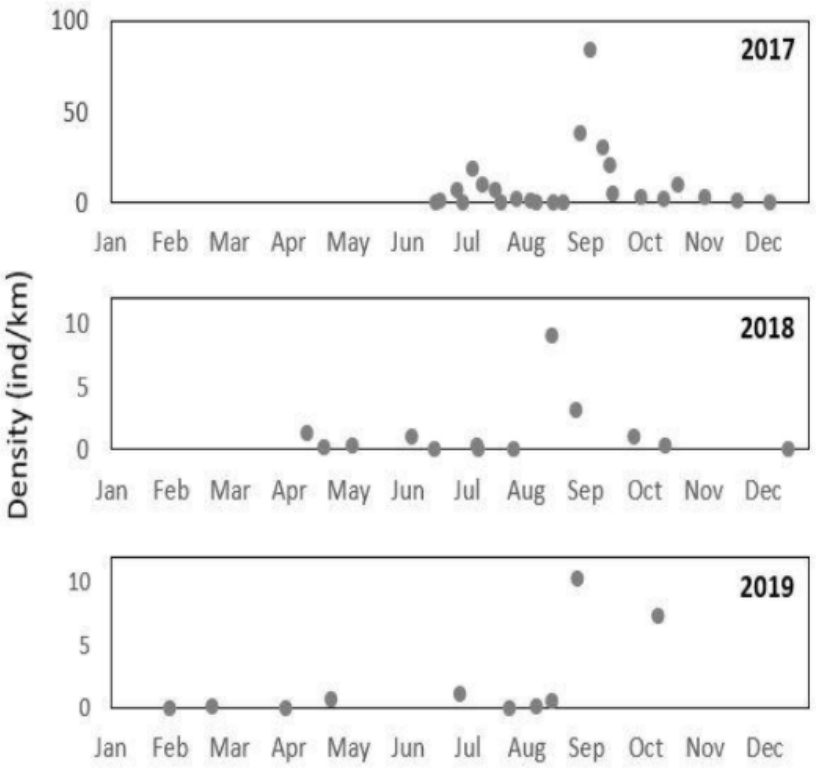


Figure 7.3 Density of neonate blackchin guitarfish per km of visual survey at Ma'agan Michael area, between June 2017 and October 2019.

This data is available for download at:

https://med-lter.haifa.ac.il/index.php/en/data-base/marine-top-predators/guitarfish-survey

7.c. Acoustic surveying and spatial distribution of coastal dolphin populations

Short-beaked common dolphins (*Delphinus delphis*): a mother and pup off the coast of Ashkelon (credit Dr Aviad Scheinin).

The two main coastal species found near the Israeli coast are *Tursiops truncatus* (Tt) and *Delphinus delphis* (Dd). Tt is found along the entire coastline (approx. 200 km, running north to south) but has not been observed in the north in the last 6 years. Dd, on the other hand, has been present since 2009, mainly south of Ashdod, and typically in groups of 15-20 individuals. Prior to 2009, these pods weren't present and only individual Dd were sighted on occasion. It is evident that these two species differ in distribution, though the environmental variables affecting the extent of their habitat is currently unknown. The MKMRS has been collaborating with the Israel Marine Mammal Research and Assistance Center (IMMRAC) in the collection and analysis of observation data from the past two decades.

Preliminary results

C-PODs and F-PODs (the more advanced device with higher detection capabilities) that monitor the presence and activity of dolphins, porpoises and other toothed whales were anchored in different sampling points along the Israeli coast from Achziv in the north down to Ashkelon in the south (Figure 7.4).

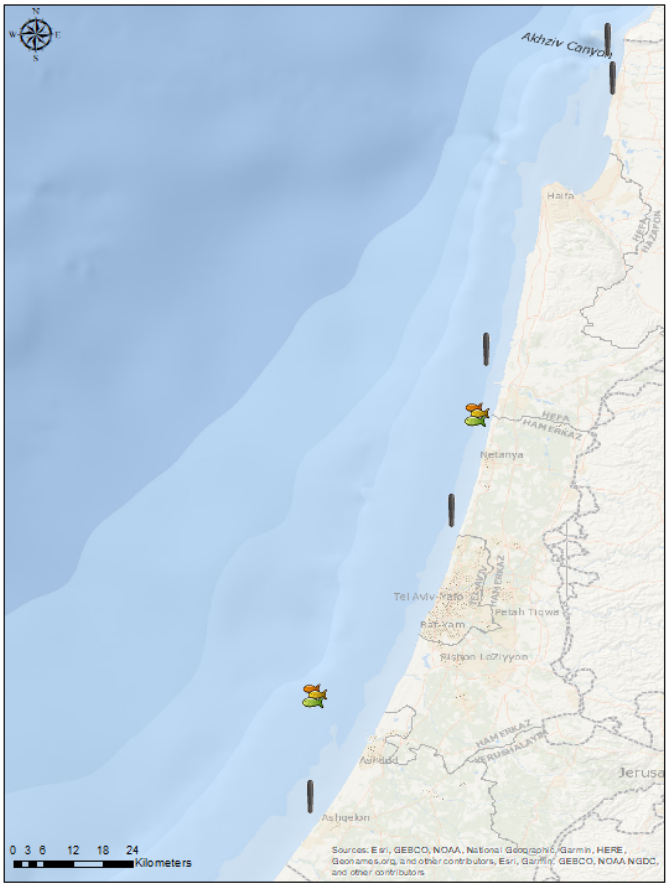


Figure 7.4. Map showing the location of the C/F-pod sampling points at rocky reefs (line) and fish farms (fish symbols).

Dolphin presence has been monitored from December 2018 until now, as can be seen in Figure 7.5.

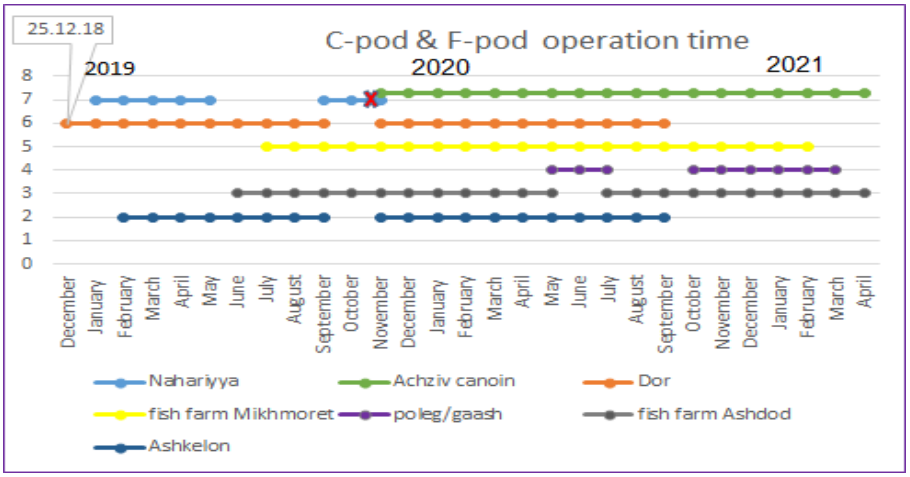


Figure 7.5: C/F-pod operation time. X - represents the change in location from Nahariya to Achziv.

DPM - Detected Positive Minutes (a minute in time containing at least one train of sound or a part of a

train produced by a dolphin) were summarised in relation to the operation time in each location (**%DPD - DPM/ minutes on*100).** Dolphins were present in all sampling points and the %DPM in each point was ranging between Nahariya and Achziv 0-0.18 % DPM, Dor 0-0.5% DPM, Hasharon 0.01-0.3% DPM and Ashkelon 0.04- 0.77% DPM. High presence of dolphins was detected in Ashdod and Michmoret fish farms with %DPM ranging between 1-49%DPM and 0.4-7%DPM, respectively. These results point to the dolphins' interest in the fish farms that are serving as hotspots.

The click parameters of the dolphins in each location were analysed and the percentage of the number of clicks per second was counted. More than 100 clicks per second (clicks/s>100) is considered as a hunting mode. The results point out that the Achziv submarine canyon is an area with lots of hunting opportunities with a mean number of clicks higher than 80 clicks/s and more than 30% of dolphins clicks detections were above 100 clicks/s. The fish farms in Ashdod and Michmoret exhibited a mean of 108 and 76 click/s and of 23.5 and 18.8 % of the number of clicks/s above 100. In all other locations including Nahariya, Dor, Hashron and Ashkelon the %clicks/s>100 was around 10% (Table 7.2).

Table 7.2. Table showing the mean number of clicks/s of dolphins click trains and % of click/s higher than 100 in each location.

	Mean # of click/s	% click/s >100
Nahariya	53.3	10
Achziv	84.39	33
Dor	52.11	12
Hasharon	42.894	9.1
Fish farm Mikhmoret	76	18.79
Fish farm Ashdod	108	23.58
Ashkelon	71.96	10

This data is available for download at:
https://med-lter.haifa.ac.il/index.php/en/data-base/marine-top-predators/dolphin-acoustic-survey

7.d. Bluefin tuna and pelagic megafauna

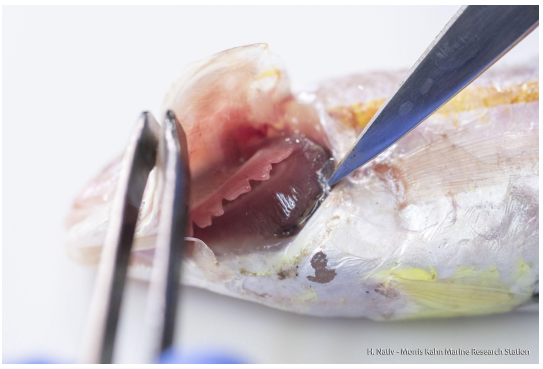
The MKMRS team has been conducting a study on the eastern Mediterranean population of Atlantic

bluefin tuna (BFT, *Thunnus thynnus*) in collaboration with onboard longline vessels of the offshore fishing fleet of Israel. This is an important species due to its biological function as an apex predator and its targeted status across the globe due to its high commercial value.

To date, 11 satellite archival tags have been deployed in collaboration with Prof. Barbara Block of Stanford University's Hopkins Marine Station and analyses are ongoing. Other sampled species included blue sharks, bigeye thresher sharks, makos and swordfish. The first attempt to fit a bigeye thresher shark with a satellite archival (PSAT) tag in this region was conducted in June 2019. This species is listed by the International Union for Conservation of Nature (IUCN) as 'Endangered' in the Mediterranean Sea and is listed as 'Vulnerable,' globally. In addition, we were able to retrieve two of the surfaced tuna tags following their programmed release and obtained the archived data at an extremely high resolution. Finally, the most recent tag to have surfaced was deployed for 465 days – a world record for the longest deployment of a tag deployed on this species. This project highlights the importance of a continuous tagging initiative in the eastern Mediterranean Sea and warrants a greater investment in the transfer of scientific knowledge to this region.

8. Marine pathology program

Scientific supervisor: Dr Dan Morick



Gill sampling of the goldband goatfish (*Upeneus moluccensis*)

Table 8.1 Summary of activity for the marine pathology program, 2016-2020

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Annual collection of 300 fish from 10 different species, conducting necropsies and sub-sampling tissues.	Molecular screening of fish for 10 different marine pathogens.	Limited staff to execute planned program	Preliminary prevalence data for five different marine pathogens from 400 fish individuals	Using New Generation Sequencing (NGS) for screening multiple pathogens (see below). Analysing pooled samples instead of individual samples for better coverage.
Annual collection of 300 fish from five different species, conducting necropsies and sub-sampling tissues (2018-2020).	Simultaneous identification of three potential fish bacterial pathogens: *Photobacterium damselae, Vibrio harveyi* and *Streptococcus iniae.*	None	20% relative abundance for classification of samples positive for *Vibrio harvei, Photobacterium damselae* and *Streptococcus iniae* as potential pathogenic bacteria-infected samples. Results are detailed in Table 8.2 and 8.3	Larger number of samples. Improvement of the tool for screening viruses, bacteria and protozoa.

Table 8.1 Summary of activity for the marine pathology program, 2016-2020- CONTINUED

Data collected	Planned data	Problems encountered	Main results	Suggested improvements
Stranded marine mammal necropsy data and tissue bank	Tissue bank and necropsy data	None	First EMS report of parasite *Toxoplasma gondii* infection as well as first detection of important zoonotic pathogens in stranded dolphins	None
Blood and serum from sharks, guitarfish and sea turtles.	Blood sampling from 40 sharks, 40 guitarfish and 40 sea turtles	Technical problems with blood handling and analysis.	Serum biochemistry results are in analysis.	Consulting a veterinary clinical pathology specialist

Understanding disease pathways, the mechanisms behind transmission, and having a baseline knowledge of the prevalence of these pathogens and viruses is crucial to the mariculture of fish. Our pathology program helps to develop methods for disease diagnosis and control, to be able to assist managers in making better decisions regarding the production yield, farm expansion, and impact of the operations. Data and basic visualisation is provided at: http://med-lter.haifa.ac.il/index.php/data-base/fish-pathogens

Methods

NGS uses high-throughput sequencing of partial 16S rRNA gene fragments. Bacterial profiles were compared by non-metric multidimensional scaling analysis (NMDS) using the Bray-Curtis distance metric. Similarly, NGS is currently under development to also detect viral and parasitic pathogens.

Results

A few studies are already in the process of finalising the data, and we are starting to have a partial understanding of pathogen abundance in the local marine environment. The Marine Pathology laboratory has focused on identifying the most suitable screening tool for evaluating marine animal

health for our region during the first four years of its activity. This has resulted in several publications, such as the below study by Meron et al. (2020). The latter study describes our preliminary results, and will serve as a basis for our routine long-term study to evaluate pathogen abundance in wild marine fish, using the 16S rRNA next-generation sequencing (NGS) screening tool for simultaneous identification of three potential fish bacterial pathogens.

a

	Fish sp.	Kidney (n)	Liver (n)	Total (n)
	Mullus barbatus	5	4	9
	Mullus surmuletus	15	12	27
	Nemipterus randalli	10	6	16
	Sardinella aurita	17	17	34
	Saurida lessepsianus	8	8	16

b

Figure 8.1 Summary of fish species and number of samples collected (kidney and liver tissues)(a). Fish illustrations were taken from http://www.fao.org/fishery/en. Israeli shoreline map includes the four sampling sites (Acre, Kishon (near Haifa), Jaffa and Ashdod) marked with asterisks (b).

Although fish were apparently healthy, 16S rRNA NGS screening identified three potential fish bacterial pathogens: *Photobacterium damselae*, *Vibrio harveyi* and *Streptococcus iniae*. Based on the distribution patterns and relative abundance, 16 samples were classified as Potential Pathogenic Bacteria-Infected Samples (PPBIS). PPBIS prevalence was significantly higher in kidneys than in liver samples. Significant differences were also observed between fish species and sites.

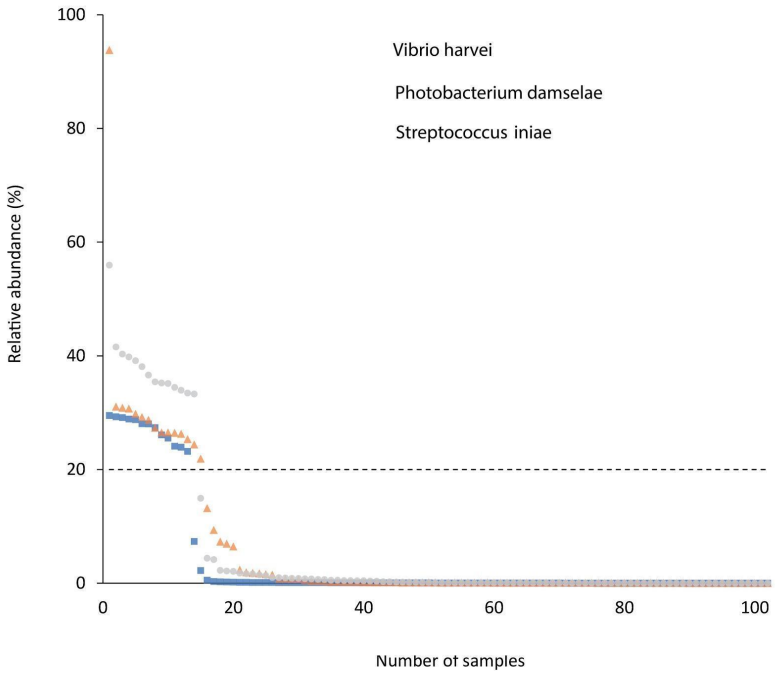


Figure 8.2. Relative abundance of pathogens in range order across all samples. Based on the distribution, we determined a threshold of 20% relative abundance (dashed line) for the classification of samples positive for Vibrio harvei, Photobacterium damselae *and* Streptococcus iniae *as potential pathogenic bacteria-infected samples (PPBIS).*

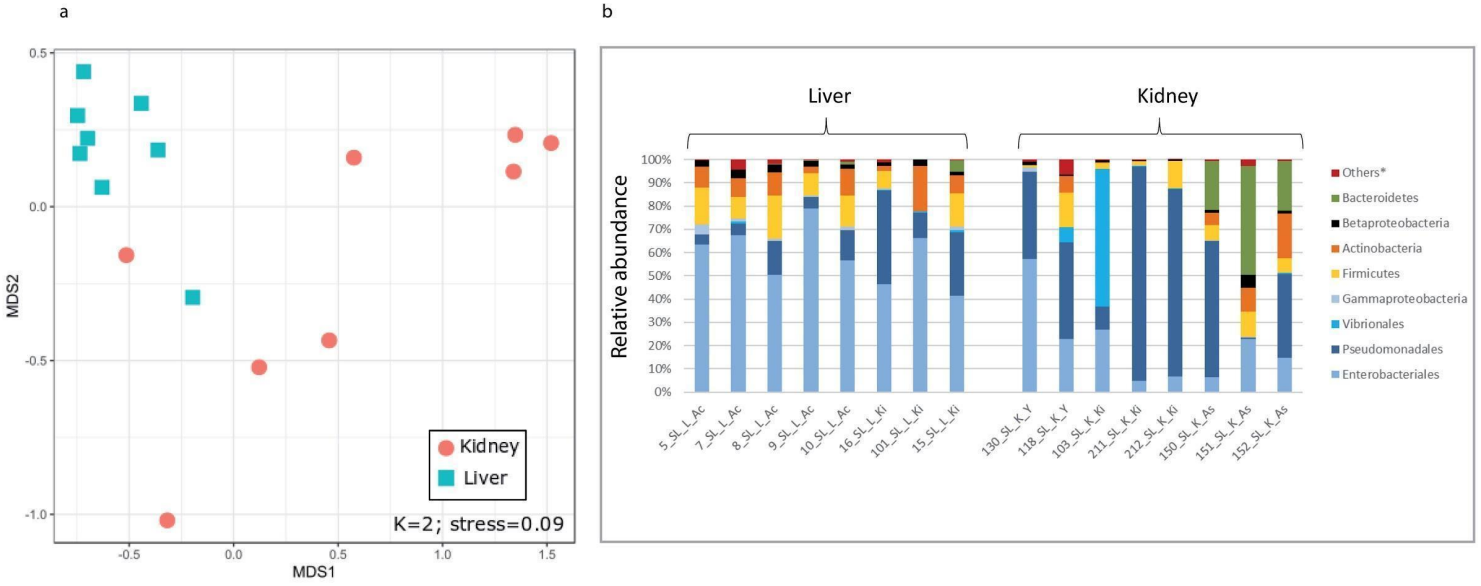


Figure 8.3. Composition and diversity of fish species and organ bacterial communities. (a) Variation in prevalence of bacterial orders across five fish species: Saurida lessepsianus (SL), Sardinella aurita

(SA), Nemipterus randalli (NR), Mullus surmuletus (MS) and Mullus barbatus (MB*) and two organs: kidney and liver.*

Table 8.2. Prevalence of potential pathogenic bacteria-infected samples (PPBIS) in kidney tissue samples. across five fish species: Saurida lessepsianus (SL), Sardinella aurita (SA), Nemipterus randalli (NR), Mullus surmuletus (MS) and Mullus barbatus (MB*).*

Potenial pathogen/Species	MB	NR	MS	SA	SL	Total
Streptococcus iniae	0	0	1	0	0	1
Photobacterium damselae	0	0	0	2	0	2
Vibrio harveyi	0	0	0	0	0	0
All 3 pathogens	3	6	2	1	0	12
No. of Kidney samples	4	10	15	16	8	53
PPBIS in Kidney	75%	60%	20%	19%	0%	23%

Table 8.3. Prevalence of potential pathogenic bacteria-infected samples (PPBIS) in samples among sites.

	Ashdod	Kishon	Yafo	Acre	Total
PPBIS	0	4	7	5	16
No. of samples	5	45	36	16	102
% PPBIS	0	9%	19%	31%	16%

Currently, we are in the process of applying this tool to stored tissue samples from our tissue bank and we hope to have the results of the last five years completed by next year.

Adding gills as another target organ for fish pathogens

The presence of pathogenic bacteria in kidneys and livers may be an indication of an immune system fault, the sign of a disease or the remains of one, as these are internal organs which do not come in direct contact with the external environment (Gorrisen and Flik, 2016). Therefore, as indicated above, kidneys were initially targeted (with liver as a secondary organ, for inter-organ comparison), and our earlier study found interesting results (Meron et al., 2020). The following study aimed, at first, to follow up on these results, but with the addition of the gills to improve the scope of the research.

Gills are an important gateway into the fish body. They are a mucosal organ involved in gas exchange, osmoregulation, acid-base regulation and excretion of nitrogenous waste, making them vital to maintaining systemic homeostasis in the face of changing internal (e.g., acidosis) and environmental (e.g., salinity) conditions (Evans et al., 2005). Gills express immune responses at both independent and systemic levels, and are not necessarily affected by pathogens entering the body. Moreover, it has been shown that mucosal vaccines are simpler and more cost effective than traditional delivery systems (Koppang et al., 2015). It should be noted that being in direct interaction with the surrounding waters

entails that the microbial communities within gills would reflect to some extent those of the water body (Kuang et al., 2020) – including the presence of pathogens – which raises an intriguing opportunity to prove the viability of using gills as a proxy for pathogen detection in the water. This study is still in the data analysis stage, but already proven to demonstrate interesting results, as can be seen at Figure 8.4.

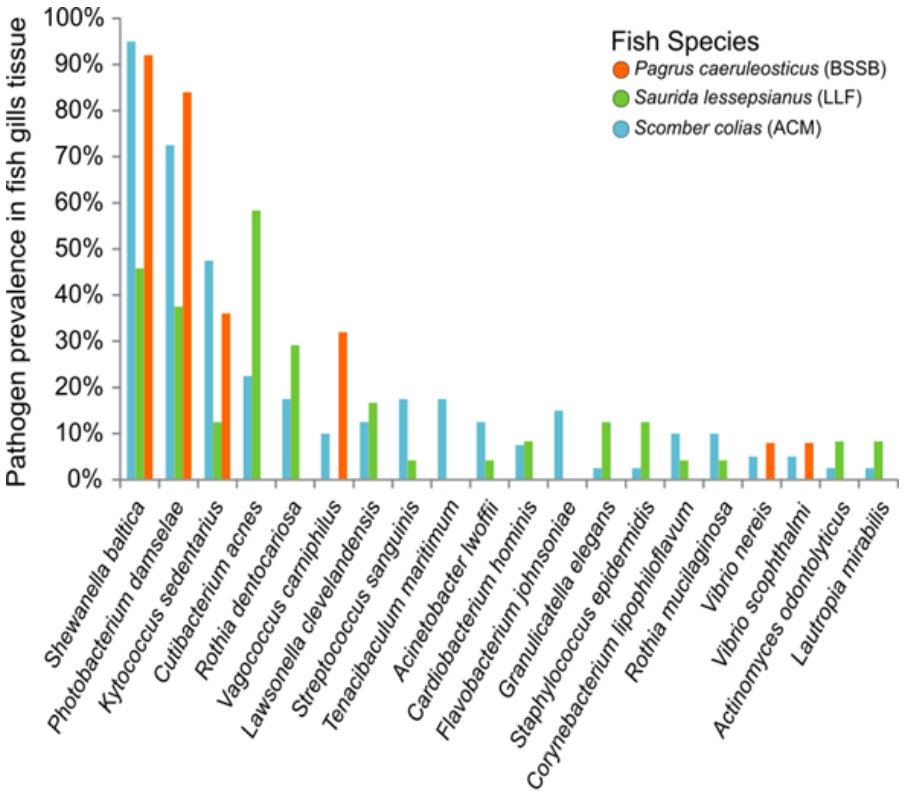


Figure 8.4. Prevalence of the 20 most commonly found pathogens in the gills of the fish sampled. The percentage (y axis) refers to the amount of samples per species found to contain these specific potential pathogens. Pathogens arranged (on the x axis) from the most (overall) prevalent (left) to the least (right).

C. Publications accepted as of 2021

1. Israel, A., Guttman, L., Shpige, M. and Neori, A., 2017. Development of polyculture and integrated multi-trophic aquaculture (IMTA) in Israel: a review. *Israeli Journal of Aquaculture-Bamidgeh*, *69*, p.20874.
2. Nobre, A.M., Valente, L.M.P. and Neori, A., 2017. A nitrogen budget model with a user-friendly interface, to assess water renewal rates and nitrogen limitation in commercial seaweed farms. *Journal of Applied Phycology*, *29*(6), pp.3039-3055.
3. Notarbartolo di Sciara G., Kerem D., Smeenk C., Rudolph P., Cesario A., Costa M., Elasar M., Feingold D., Fumagalli M., Goffman O., Hadar N., Mebrathu Y.T., Scheinin A. Cetaceans of the Red Sea. CMS Technical Series 33, 86 pp.
4. Powley, H.R., Krom, M.D. and Van Cappellen, P., 2017. Understanding the unique biogeochemistry of the Mediterranean Sea: Insights from a coupled phosphorus and nitrogen model. *Global Biogeochemical Cycles*, *31*(6), pp.1010-1031.
5. Barash, A., Pickholtz, R., Nativ, H., Malamud, S., Scheinin, A. and Tchernov, D., 2018. Seasonal arrival and feeding of injured coastal sharks at fish farms in the Eastern Mediterranean. *Journal of the Black Sea/Mediterranean Environment*, *24*(1), pp.86-90.
6. Yeruham, E., Abelson, A., Rilov, G., Ezra, D.B. and Shpigel, M., 2019. Energy budget of cultured Paracentrotus lividus under different temperatures. *Aquaculture*, *501*, pp.7-13.
7. Bigal, E., Morick, D., Scheinin, A.P., Salant, H., Berkowitz, A., King, R., Levy, Y., Melero, M., Sánchez-Vizcaíno, J.M., Goffman, O. and Hadar, N., 2018. Detection of Toxoplasma gondii in three common bottlenose dolphins (Tursiops truncatus); A first description from the Eastern Mediterranean Sea. *Veterinary parasitology*, *258*, pp.74-78.
8. Chen, N., Krom, M.D., Wu, Y., Yu, D. and Hong, H., 2018. Storm induced estuarine turbidity maxima and controls on nutrient fluxes across river-estuary-coast continuum. Science of the Total Environment, 628, pp.1108-1120.
9. Herbert, R.J., Krom, M.D., Carslaw, K.S., Stockdale, A., Mortimer, R.J.G., Benning, L.G., Pringle, K. and Browse, J., 2018. The effect of atmospheric acid processing on the global deposition of bioavailable phosphorus from dust. *Global Biogeochemical Cycles*, *32*(9), pp.1367-1385.
10. Levy, D., Belfer, Y., Osherov, E., Bigal, E., Scheinin, A.P., Nativ, H., Tchernov, D. and Treibitz, T., 2018. Automated analysis of marine video with limited data. In *Proceedings of the IEEE Conference on Computer Vision and Pattern Recognition Workshops* (pp. 1385-1393).
11. Mannocci, L., Roberts, J.J., Halpin, P.N., Authier, M., Boisseau, O., Bradai, M.N., Cañadas, A., Chicote, C., David, L., Di-Méglio, N. and Fortuna, C.M., 2018. Assessing cetacean surveys throughout the Mediterranean Sea: a gap analysis in environmental space. *Scientific reports*, *8*(1), pp.1-14.
12. Milstein, A., Levy, A., Neori, A., Harpaz, S., Shpigel, M. and Guttman, L., 2018. Water quality, ecological processes and management procedures in a periphyton biofiltration system in mariculture: A statistical analysis. *Aquaculture Research*, *49*(4), pp.1491-1503.
13. Shpigel, M. and Erez, J., 2020. Effect of diets and light regimes on calcification and somatic growth of the sea urchin Tripneustes gratilla elatensis. *Aquaculture*, *529*, p.735547.
14. Shpigel, M., Guttman, L., Ben-Ezra, D., Yu, J. and Chen, S., 2019. Is Ulva sp. able to be an efficient biofilter for mariculture effluents?. *Journal of Applied Phycology*, *31*(4), pp.2449-2459.
15. Shpigel, M., Shauli, L., Odintsov, V., Ashkenazi, N. and Ben-Ezra, D., 2018. Ulva lactuca biofilter from a land-based integrated multi trophic aquaculture (IMTA) system as a sole food source for the tropical sea urchin Tripneustes gratilla elatensis. *Aquaculture*, *496*, pp.221-231.
16. Brand, D., Edelist, D., Goffman, O., Hadar, N., Scheinin, A. and Kerem, D., 2019. Common dolphins, common in neritic waters off southern Israel, demonstrate uncommon dietary habits. *Aquatic Conservation: Marine and Freshwater Ecosystems*, *31*, pp.15-21.

17. Alexandri, T., Shamir, Z.Z., Bigal, E., Scheinin, A., Tchernov, D. and Diamant, R., 2019. Localization of acoustically tagged marine animals in under-ranked conditions. *IEEE Transactions on Mobile Computing*, *20*(3), pp.1126-1137.
18. Israel, A., Golberg, A. and Neori, A., 2020. The seaweed resources of Israel in the Eastern Mediterranean Sea. *Botanica Marina*, *63*(1), pp.85-95.
19. Guttman, L., Neori, A., Boxman, S.E., Barkan, R., Shahar, B., Tarnecki, A.M., Brennan, N.P., Main, K.L. and Shpigel, M., 2019. An integrated Ulva-periphyton biofilter for mariculture effluents: Multiple nitrogen removal kinetics. *Algal Research*, *42*, p.101586.
20. Suari, Y., Amit, T., Gilboa, M., Sade, T., Krom, M.D., Gafny, S., Topaz, T. and Yahel, G., 2019. Sandbar breaches control of the biogeochemistry of a micro-estuary. *Frontiers in Marine Science*, *6*, p.224.
21. Yeruham, E., Shpigel, M., Abelson, A. and Rilov, G., 2020. Ocean warming and tropical invaders erode the performance of a key herbivore. *Ecology*, *101*(2), p.e02925.
22. Neori, A., Guttman, L., Israel, A. and Shpigel, M., 2019. Israeli-developed models of marine integrated multi trophic aquaculture (IMTA). *Journal of Coastal Research*, *86*(SI), pp.11-20.
23. Zirks, E., Krom, M.D., Zhu, D., Schmiedl, G. and Goodman-Tchernov, B.N., 2019. Evidence for the presence of oxygen-depleted sapropel intermediate water across the eastern mediterranean during sapropel S1. *ACS Earth and Space Chemistry*, *3*(10), pp.2287-2297.
24. Meron, D., Maor-Landaw, K., Weizman, E., Waldman Ben-Asher, H., Eyal, G., Banin, E., Loya, Y. and Levy, O., 2019. The algal symbiont modifies the transcriptome of the Scleractinian coral Euphyllia paradivisa during heat stress. *Microorganisms*, *7*(8), p.256.
25. Shamir, Z.Z., Shamir, S.Z., Becker, N., Scheinin, A. and Tchernov, D., 2019. Evidence of the impacts of emerging shark tourism in the Mediterranean. *Ocean & Coastal Management*, *178*, p.104847.
26. Thompson, J., Poulton, S.W., Guilbaud, R., Doyle, K.A., Reid, S. and Krom, M.D., 2019. Development of a modified SEDEX phosphorus speciation method for ancient rocks and modern iron-rich sediments. *Chemical Geology*, *524*, pp.383-393.
27. Shkury, G., Bud, J., Zuriel, Y., Scheinin, A. and Diamant, R., 2019, June. Robust Automatic Detector And Feature Extractor For Dolphin Whistles. In *OCEANS 2019-Marseille* (pp. 1-7). IEEE.
28. Gutner-Hoch, E., Martins, R., Maia, F., Oliveira, T., Shpigel, M., Weis, M., Tedim, J. and Benayahu, Y., 2019. Toxicity of engineered micro-and nanomaterials with antifouling properties to the brine shrimp Artemia salina and embryonic stages of the sea urchin Paracentrotus lividus. *Environmental Pollution*, *251*, pp.530-537.
29. Zemah Shamir, Z., Zemah Shamir, S., Tchernov, D., Scheinin, A. and Becker, N., 2019. Shark aggregation and tourism: opportunities and challenges of an emerging phenomenon. *International Journal of Sustainable Development & World Ecology*, *26*(5), pp.406-414.
30. Diamant, R., Kipnis, D., Bigal, E., Scheinin, A., Tchernov, D. and Pinchasi, A., 2019. An active acoustic track-before-detect approach for finding underwater mobile targets. *IEEE Journal of Selected Topics in Signal Processing*, *13*(1), pp.104-119.
31. Xiong, Y., Guilbaud, R., Peacock, C.L., Cox, R.P., Canfield, D.E., Krom, M.D. and Poulton, S.W., 2019. Phosphorus cycling in Lake Cadagno, Switzerland: A low sulfate euxinic ocean analogue. *Geochimica et Cosmochimica Acta*, *251*, pp.116-135.
32. Yu, D., Chen, N., Krom, M.D., Lin, J., Cheng, P., Yu, F., Guo, W., Hong, H. and Gao, X., 2019. Understanding how estuarine hydrology controls ammonium and other inorganic nitrogen concentrations and fluxes through the subtropical Jiulong River Estuary, SE China under baseflow and flood-affected conditions. *Biogeochemistry*, *142*(3), pp.443-466.
33. Berzak, R., Scheinin, A., Davidovich, N., Regev, Y., Diga, R., Tchernov, D. and Morick, D., 2019. Prevalence of nervous necrosis virus (NNV) and Streptococcus species in wild marine fish and crustaceans from the Levantine Basin, Mediterranean Sea. *Diseases of aquatic organisms*, *133*(1), pp.7-17.
34. Kroeger, L.A., Tsemel, A., Tchernov, D. and Ayalon, O., 2019. A theoretical approach concerning nutrient

emissions to inform monitoring and management of mariculture farms. *Mar. Sci*, *2*, pp.125-133.

35. Shahar, B., Shpigel, M., Barkan, R., Masasa, M., Neori, A., Chernov, H., Salomon, E., Kiflawi, M. and Guttman, L., 2020. Changes in metabolism, growth and nutrient uptake of Ulva fasciata (Chlorophyta) in response to nitrogen source. *Algal Research*, *46*, p.101781.

36. Martinez, S., Lalzer, M., Shemesh, E., Einbinder, S., Tchernov, B.G. and Tchernov, D., 2020. Effect of different derivatization protocols on the calculation of trophic position using amino acids compound-specific stable isotopes. *bioRxiv*.

37. Martinez, S., Kolodny, Y., Shemesh, E., Scucchia, F., Nevo, R., Levin-Zaidman, S., Paltiel, Y., Keren, N., Tchernov, D. and Mass, T., 2020. Energy sources of the depth-generalist mixotrophic coral Stylophora pistillata. *Frontiers in Marine Science*, *7*.

38. Wosnick, N., Chaves, A.P., Niella, Y.V., Takatsuka, V., Hazin, F.H.V., Nunes, J.L.S. and Morick, D., 2020. Physiological impairment as a result of bile accumulation in an apex predator, the tiger shark (Galeocerdo cuvier Péron & Lesueur, 1822). *Animals*, *10*(11), p.2030.

39. Genin, A., Levy, L., Sharon, G., Raitsos, D.E. and Diamant, A., 2020. Rapid onsets of warming events trigger mass mortality of coral reef fish. *Proceedings of the National Academy of Sciences*, *117*(41), pp.25378-25385.

40. Heckman, T.I., Griffin, M.J., Camus, A.C., LaFrentz, B.R., Morick, D., Smirnov, R., Ofek, T. and Soto, E., 2020. Multilocus sequence analysis of diverse Streptococcus iniae isolates indicates an underlying genetic basis for phenotypic heterogeneity. *Diseases of aquatic organisms*, *141*, pp.53-69.

41. Neori and Agami. 2020. The Synergy between Herbicides, Oil and Solvents in Bio-Toxicity to Phaeodactylum tricornutumBohlin-Implications forInterpretation of Field SampleData. Expert Opin Environ Biol 2020, 9:2. DOI: 10.37532/eoeb.2020.9(2).161

42. Davidovich, N., Pretto, T., Sharon, G., Zilberg, D., Blum, S.E., Baider, Z., Edery, N., Morick, D., Grossman, R., Kaidar-Shwartz, H. and Dveyrin, Z., 2020. Cutaneous appearance of mycobacteriosis caused by Mycobacterium marinum, affecting gilthead seabream (Sparus aurata) cultured in recirculating aquaculture systems. *Aquaculture*, *528*, p.735507.

43. Pardo JCF, Ramon D, Stefanelli-Silva G, Elegbede I, Lima LS and Principe SC. 2020. Advancing Through the Pandemic From the Perspective of Marine Graduate Researchers: Challenges, Solutions, and Opportunities. Front. Mar. Sci. 7:528. doi: 0.3389/fmars.2020.0052

44. Livne, L., Grossowicz, M., Tchernov, D. and Ayalon, O., 2020. Predicting Impacts of Offshore Monoculture Farm Expansion in Ultra-Oligotrophic Waters of the Levantine Basin. *Frontiers in Marine Science*, *7*, p.556.

45. Casari, P., Lampe, L., Martinez, S. and Diamant, R., 2020. ThreatDetect: An Autonomous Platform to secure Marine Infrastructures. In *Advanced Technologies for Security Applications* (pp. 271-281). Springer, Dordrecht.

46. Chang, Z.Q., Neori, A., He, Y.Y., Li, J.T., Qiao, L., Preston, S.I., Liu, P. and Li, J., 2020. Development and current state of seawater shrimp farming, with an emphasis on integrated multi-trophic pond aquaculture farms, in China–a review. *Reviews in Aquaculture*, *12*(4), pp.2544-2558.

47. Shpigel, M. and Erez, J., 2020. Effect of diets and light regimes on calcification and somatic growth of the sea urchin Tripneustes gratilla elatensis. *Aquaculture*, *529*, p.735547.

48. Neori, A., Bronfman, Y., Van Rijn, J., Guttman, L., Krupnik, N., Shpigel, M., Samocha, T.M., Davis, D.A., Qiu, X., Abelin, P. and Israel, Á., 2020. The suitability of Ulva fasciata, Ulva compressa, and Hypnea musciformis for production in an outdoor spray cultivation system, with respect to biomass yield and protein content. *Journal of Applied Phycology*, *32*, pp.3183-3197.

49. Regev, Y., Davidovich, N., Berzak, R., Lau, S.C., Scheinin, A.P., Tchernov, D. and Morick, D., 2020. Molecular identification and characterization of Vibrio species and Mycobacterium species in wild and cultured marine fish from the Eastern Mediterranean Sea. *Microorganisms*, *8*(6), p.863.

50. Lampert, Y., Berzak, R., Davidovich, N., Diamant, A., Stern, N., Scheinin, A.P., Tchernov, D. and Morick, D., 2020. Indigenous versus Lessepsian Hosts: Nervous Necrosis Virus (NNV) in Eastern Mediterranean Sea Fish. *Viruses*, *12*(4), p.430.

51. Meron, D., Davidovich, N., Ofek-Lalzar, M., Berzak, R., Scheinin, A., Regev, Y., Diga, R., Tchernov, D. and Morick, D., 2020. Specific pathogens and microbial abundance within liver and kidney tissues of wild marine fish from the Eastern Mediterranean Sea. *Microbial biotechnology*, *13*(3), pp.770-780.

52. Israel, A., Golberg, A. and Neori, A., 2020. The seaweed resources of Israel in the Eastern Mediterranean Sea. *Botanica Marina*, *63*(1), pp.85-95.

53. Meron, D., Maor-Landaw, K., Eyal, G., Elifantz, H., Banin, E., Loya, Y. and Levy, O., 2020. The Complexity of the Holobiont in the Red Sea Coral Euphyllia paradivisa under Heat Stress. *Microorganisms*, *8*(3), p.372.

54. Grossowicz, M., Shemesh, E., Martinez, S., Bialik, O.M., Benayahu, Y. and Tchernov, D., 2020, February. New evidence for colonization of the migrant octocoral Melithaea erythraea in the Eastern Mediterranean. In *Ocean Sciences Meeting 2020*. AGU.

55. Morick, D., Davidovich, N., Bigal, E., Rosenbluth, E., Bouznach, A., Rokney, A., Ron, M., Wosnick, N., Tchernov, D. and Scheinin, A.P., 2020. Fatal Infection in a Wild Sandbar Shark (Carcharhinus plumbeus), Caused by Streptococcus agalactiae, Type Ia-ST7. *Animals*, *10*(2), p.284.

56. Knowler, D., Chopin, T., Martínez-Espiñeira, R., Neori, A., Nobre, A., Noce, A. and Reid, G., 2020. The economics of Integrated Multi-Trophic Aquaculture: where are we now and where do we need to go?. *Reviews in Aquaculture*, *12*(3), pp.1579-1594.

57. Scucchia, F., Nativ, H., Neder, M., Goodbody-Gringley, G. and Mass, T., 2020. Physiological characteristics of Stylophora pistillata larvae across a depth gradient. *Frontiers in Marine Science*, *7*, p.13.

58. Malik, A., Einbinder, S., Martinez, S., Tchernov, D., Haviv, S., Almuly, R., Zaslansky, P., Polishchuk, I., Pokroy, B., Stolarski, J. and Mass, T., 2021. Molecular and skeletal fingerprints of scleractinian coral biomineralization: from the sea surface to mesophotic depths. *Acta biomaterialia*, *120*, pp.263-276.

59. Liu, J., Krom, M.D., Ran, X., Zang, J., Liu, J., Yao, Q. and Yu, Z., 2020. Sedimentary phosphorus cycling and budget in the seasonally hypoxic coastal area of Changjiang Estuary. *Science of The Total Environment*, *713*, p.136389.

60. Shahar, B., Shpigel, M., Barkan, R., Masasa, M., Neori, A., Chernov, H., Salomon, E., Kiflawi, M. and Guttman, L., 2020. Changes in metabolism, growth and nutrient uptake of Ulva fasciata (Chlorophyta) in response to nitrogen source. *Algal Research*, *46*, p.101781.

61. Drake, J., Malik, A., Popovits, Y., Yosef, O., Shamesh, E., Stolarski, J., Tchernov, D., Sher, D. and Mass, T., 2021. Physiological and transcriptomic variability indicative of differences in key functions within a single coral colony. *bioRxiv*.

62. Gefen-Treves, S., Bartholomäus, A., Horn, F., Zaborowski, A.B., Tchernov, D., Wagner, D., Oren, A. and Kaplan, A., 2021. The Microbiome Associated with the Reef Builder Neogoniolithon sp. in the Eastern Mediterranean. *Microorganisms*, *9*(7), p.1374.

63. Yuval, M., Alonso, I., Eyal, G., Tchernov, D., Loya, Y., Murillo, A.C. and Treibitz, T., 2021. Repeatable semantic reef-mapping through photogrammetry and label-augmentation. *Remote Sensing*, *13*(4), p.659.

64. Rearte, T.A., Celis-Plá, P.S., Neori, A., Masojídek, J., Torzillo, G., Gómez-Serrano, C., Benavides, A.M.S., Álvarez-Gómez, F., Abdala-Díaz, R.T., Ranglová, K. and Caporgno, M., 2021. Photosynthetic performance of Chlorella vulgaris R117 mass culture is moderated by diurnal oxygen gradients in an outdoor thin layer cascade. *Algal Research*, *54*, p.102176.

65. Zirks, E., Krom, M., Schmiedl, G., Katz, T., Xiong, Y., Alcott, L.J., Poulton, S.W. and Goodman-Tchernov, B., 2021. Redox evolution and the development of oxygen minimum zones in the Eastern Mediterranean Levantine basin during the early Holocene. *Geochimica et Cosmochimica Acta*, *297*, pp.82-100.

66. Yuan, X., Krom, M.D., Zhang, M. and Chen, N., 2021. Human disturbance on phosphorus sources, processes and riverine export in a subtropical watershed. *Science of The Total Environment*, *769*, p.144658.

67. Ferrier-Pagès, C., Martinez, S., Grover, R., Cybulski, J., Shemesh, E. and Tchernov, D., 2021. Tracing the Trophic Plasticity of the Coral–Dinoflagellate Symbiosis Using Amino Acid Compound-Specific Stable Isotope Analysis. *Microorganisms*, *9*(1), p.182.

D. Bibliography

Ben-Ezra, Tal, Anat. Tsemel, Ilana Berman-Frank, Barak Herut, Yoav Lehahn, Eyal Rahav, Tom Reich, Tron Frede Thingstad, and Daniel Sher. 2021. "Seasonal Nutrient Dynamics in the P Depleted Eastern Mediterranean Sea." Deep Sea Research Part I: Vol 176 Vol 176 DOI: 10.1016/j.dsr.2021.103607

Berman, T., Y. Azov, A. Schneller, P. Walline, and D.W. W Townsend. 1986. "Extent, Transparency and Phytoplankton Distribution of the Neritic Water Overlying the Israeli Coastal Shelf." Oceanologica Acta 4: 439–47. http://archimer.ifremer.fr/doc/00267/37818/35899.pdf

Bialik, Or M., and Guy Sisma-Ventura. 2016. "Proxy-Based Reconstruction of Surface Water Acidification and Carbonate Saturation of the Levant Sea during the Anthropocene." Anthropocene 16: 42–53. https://doi.org/10.1016/j.ancene.2016.08.001

Brokovich, E., Einbinder, S., Shashar, N., Kiflawi, M., & Kark, S. (2008). Descending to the twilight-zone: changes in coral reef fish assemblages along a depth gradient down to 65 m. Marine Ecology Progress Series, 371, 253–262. http://www.jstor.org/stable/24872704

Duer, D. M. 1993. Biological Criteria, Environmental health and estuarine macrobenthic community structure. Mar. Pollut. Bull. Boesch Rosenb. 26, 249–257

Edelist, D., A. Scheinin, O. Sonin, J. Shapiro, P. Salameh, G. Rilov, Y Benayahu, D. Schultz, and D. Zeller. 2013. "Israel: Reconstructed Estimates of Total Fisheries Removals in the Mediterranean, 1950–2010." Acta Adriatica, no. 54: 256–64.

Edelist, D.. 2012. "Fishery Management and Marine Invasion in Israel." Mount Carmel: University of Haifa

Evans, D. H., Piermarini, P. M., and Choe, K. P. 2005. The Multifunctional Fish Gill: Dominant Site of Gas Exchange, Osmoregulation, Acid-Base Regulation, and Excretion of Nitrogenous Waste. Physiol. Rev. 85, 97–177. doi:10.1152/physrev.00050.2003

Fricke, R., D. Golani, and B. Appelbaum-Golani. 2017. "Arnoglossus Nigrofilamentosus n. Sp., a New Species of Flounder (Teleostei: Bothidae) from off the Mediterranean Coast of Israel, Probably a New Case of Lessepsian Migration." Scientia Marina 81 (4): 457. https://doi.org/10.3989/scimar.04684.07A

Gadd, G. M., & Griffiths, A. J. (1977). Microorganisms and heavy metal toxicity. Microbial Ecology, 4(4),

Galil, BS. 2008. Alien Species in the Mediterranean Sea—Which, When, Where, Why? Challenges to Marine Ecosystems. Netherlands: Springer. http://link.springer.com/chapter/10.1007/978-1-4020-8808-7_10

Gertman, I., J. Silverman, Israel Oceanographic, and Limnological Rese. 2016. "Interannual Thermohaline (1979 – 2014) and Nutrient (2002 – 2014) Dynamics in the Levantine Surface and Intermediate Water ..." Global and

Planetary Change, no. April. https://doi.org/10.1016/j.gloplacha.2016.04.001

Golani, D., B. Appelbaum-Golani, and O. Gon. 2008. "Apogon Smithi (Kotthaus, 1970) (Teleostei: Apogonidae), a Red Sea Cardinalfish Colonizing the Mediterranean Sea." Journal of Fish Biology 72 (6): 1534–38. https://doi.org/10.1111/j.1095-8649.2008.01812.x

Gorissen, M., and Flik, G. 2016. "The Endocrinology of the Stress Response in Fish," in Fish Physiology, ed. C. J. B. Carl B. Schreck, Lluis Tort, Anthony P. Farrell (Academic Press), 75–111. doi:10.1016/B978-0-12-802728-8.00003-5

Halim, Y. 1960. "Observations on the Nile Bloom of Phytoplankton in the Mediterranean." Journal Du Conseil 26 (1): 57–67.

303–317. https://doi.org/10.1007/BF02013274).

Kennish, M. J. (2002). Environmental threats and environmental future of estuaries. Environmental Conservation, 29(01), 78–107. https://doi.org/10.1017/S0376892902000061

Krom, M. D, N. Kress, and S. Brenner. 1991. "Phosphorus Limitation of Primary Productivity in the Eastern Mediterranean Sea." Limnology and Oceanography 36: 424–32

Krom, M.D., and Y. Suari. 2015. "The Eastern Mediterranean a Natural Laboratory for Studying Marine Biogeochemical Processes." Challenger Society 21 (1): 46–52

Krom, M., N. Kress, I. Berman-Frank, and E. Rahav. 2014. "Past, Present and Future Patterns in the Nutrient Chemistry of the Eastern Mediterranean." The Mediterranean Sea: Its History and Present Challenges, January, 49–68. https://doi.org/10.1007/978-94-007-6704-1_4

Koppang, E. O., Kvellestad, A., and Fischer, U. 2015. "Fish mucosal immunity: gill," in Mucosal Health in Aquaculture, eds. B. H. Beck, E. Peatman, and B. Allam (London: Academic Press), 93–133

Kuang, T., He, A., Lin, Y., Huang, X., Liu, L., and Zhou, L. 2020. Comparative analysis of microbial communities associated with the gill, gut, and habitat of two filter-feeding fish. Aquac. Reports 18, 100501. doi:10.1016/j.aqrep.2020.100501

Letessier, T. B., P. J. Bouchet, , and Jessica Meeuwig. 2017. "Sampling Mobile Oceanic Fishes and Sharks: Implications for Fisheries and Conservation Planning." Biological Reviews 92 (2): 627–46. https://doi.org/10.1111/brv.12246

Letessier, T. B., Bouchet, P. J., and Meeuwig, J. J. 2015. Sampling mobile oceanic fishes and sharks: Implications for fisheries and conservation planning. Biol. Rev. doi:10.1111/brv.12246.

Meron, D., Davidovich, N., Ofek-Lalzar, M., Berzak, R., Scheinin, A., Regev, Y., et al. 2020. Specific pathogens and microbial abundance within liver and kidney tissues of wild marine fish from the Eastern Mediterranean Sea. Microb. Biotechnol. 1–11. doi:10.1111/1751-7915.1353

Nixon, S. 2004. "The Artificial Nile." American Scientist 92 (2): 158. https://doi.org/10.1511/2004.2.158

Powley, HR, MD Krom, and P Van Cappellen. 2017. "Understanding the Unique Biogeochemistry of the Mediterranean Sea: Insights from a Coupled Phosphorus and Nitrogen Model." Global Biogeochemical Cycles 11:

1010–31

SEAL Analytical. n.d. “Ammonia in Water and Seawater.” AutoAnalyzer Applications G-327-05 (Rev 7) ———. n.d. “Nitrate and Nitrite in Water and Seawater Method-No. G-172-96.” In AutoAnalyzer Applications, Rev 17. Hamburg: SEAL Analytical.

———. n.d. “Phosphate in Water and Seawater G-170-96.” AutoAnalyzer Applications G-170-96 (Rev.3)

———. n.d. “Silicate in Water and Seawater Method-No. G-177-96.” AutoAnalyzer Applications G-177-96 (Rev.11

Seymour, J. R. 2014. A sea of microbes: the diversity and activity of marine microorganisms. Microbiol. Aust. 35, 183 Conservation, 29(01), 78–107. https://doi.org/10.1017/S0376892902000061

Shamir, Z.Z., Shamir, S.Z., Becker, N., Scheinin, A. and Tchernov, D., 2019. Evidence of the impacts of emerging shark tourism in the Mediterranean. Ocean & Coastal Management, 178, p.104847.

Suari, Y., and S. Brenner. 2015. “Decadal Biogeochemical History of the South East Levantine Basin: Simulations of the River Nile Regimes.” Journal of Marine Systems 148: 112–21. http://www.sciencedirect.com/science/article/pii/S0924796315000263.

Thingstad, TF, MD Krom, and RFC Mantoura. 2005. “Nature of Phosphorus Limitation in the Ultraoligotrophic Eastern Mediterranean.” Science. http://www.sciencemag.org/content/309/5737/1068.short.

Tsagaraki, Tatiana M., Barak Herut, Eyal Rahav, Ilana R. Berman Frank, Anastasia Tsiola, Manolis Tsapakis, Antonia Giannakourou, et al. 2017. “Atmospheric Deposition Effects on Plankton Communities in the Eastern Mediterranean: A Mesocosm Experimental Approach.” Frontiers in Marine Science 4 (JUL). https://doi.org/10.3389/fmars.2017.00210.

Zirks, E., M. Krom, G. Schmiedl, T. Katz, Y. Xiong, L. J. Alcott, S. W. Poulton, and B. Goodman-Tchernov. 2021. “Redox Evolution and the Development of Oxygen Minimum Zones in the Eastern Mediterranean Levantine Basin during the Early Holocene.” Geochimica et Cosmochimica Acta 297 (March): 82–100.