

# Abstracts

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ABSTRACTS

ORAL PRESENTATIONS

# A productivity bottleneck in the Baltic herring (Clupea harengus membras): early life-history processes and recruitment variability

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Exogenous anomalies induced by contemporary climate change may severely impact dynamics of early life stages of fish. Here, we modelled how growth rate and abundance of postflexion larvae, and recruitment of Baltic spring-spawning herring (Clupea harengus membras) in the Pärnu Bay, Gulf of Riga (GoR) may respond to shifting climate variables. Higher larval growth rates were aligned with later seasonal emergence of yolk-sac larvae, while lower abundance of postflexion larvae occurred in years of earlier seasonal seawater warming. Cooler temperatures (<16 °C) in spring expanded the optimal thermal window for first-feeding herring larvae, attributable to the absence of early seasonal water temperature warming. Higher recruitment levels emerged in years of seasonally delayed warming and were associated with higher abundance of postflexion larvae. In recent decades, the trend towards earlier warming of the Baltic Sea in spring threatens to create a bottleneck to successful recruitment of herring. The existing paradigm that abundant Baltic herring year-classes occur only in the years following mild winters no longer stands as environmental conditions undergo rapid change. The relative contribution of Pärnu Bay larval nursery areas to recruitment has diminished as the suitable thermal window has been dramatically reduced in recent decades. Evolving thermal dynamics in the GoR have developed relatively recently and in future present a bottleneck for herring production.

Why are sand lance embryos so sensitive to future oceanic CO<sub>2</sub> conditions?

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Rising oceanic pCO<sub>2</sub> levels could affect many traits in fish early life stages, but only few species to date have shown direct CO<sub>2</sub>-induced survival reductions. This might partly be because species from less CO<sub>2</sub>variable, offshore environments in higher latitudes are currently underrepresented in the literature. We conducted new experimental work on northern sand lance (Ammodytes dubius), a keystone forage fish on offshore Northwest Atlantic sand banks, which was recently suggested to be highly CO<sub>2</sub>-sensitive. In two complementary trials, we produced embryos from wild, Gulf of Maine (GoM) spawners and reared them at several pCO<sub>2</sub> levels (~400 – 2,000  $\mu$ atm) in combination with static (6, 7, 10°C) and dynamic  $(10 \rightarrow 5^{\circ}C)$  temperature treatments. Again, we consistently observed large, CO<sub>2</sub>-induced reductions in hatching success (-23% at 1,000 µatm, -61% at ~2,000 µatm), and the effects were temperatureindependent. To distinguish pCO<sub>2</sub> effects during development from potential impacts on hatching itself, some embryos were switched between high and control pCO<sub>2</sub> treatments just prior to hatch. This indeed altered hatching patterns consistent with the CO<sub>2</sub>-impaired hatching hypothesis. High CO<sub>2</sub> also delayed the day of first hatch in one trial and peak hatch in the other, where later-hatched larvae were of similar size but with progressively less endogenous energy reserves. For context, we extracted seasonal  $pCO_2$ projections for Stellwagen Bank (GoM) from regional ensemble simulations, which indicated a CO2induced reduction in sand lance hatching success to 71% of contemporary levels by 2100. The species' unusual CO<sub>2</sub> sensitivity has large ecological and scientific ramifications that warrant future in-depth research.

Changing communities: Investigating microbial community dynamics during early life-history transitions in the chemosymbiotic, deep-sea mussel *Gigantidas childressi* 

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Marine invertebrates form specific associations with bacterial communities that are different from their environment, change throughout their development, and shape evolutionary and ecological processes. The bathymodiolin (Mytilidae) mussel Gigantidas childressi lives at deep-sea methane seeps and relies on methanotrophic endosymbionts for nutrition. Its larval life, however, is spent feeding on plankton in the water column. Upon metamorphosis at a seep habitat, methanotrophic bacteria colonize gill cells and the juvenile mussel switches to symbiont-derived energy. We examined the microbiomes of G. childressi during these developmental transitions by sequencing the V3/V4 regions of the 16S rRNA gene in individual larvae and juveniles. Diversity was relatively low, and our results show strong evidence for the reorganization of the microbiome based on stage-specific shifts in habitat and nutritional mode. In larvae, the microbiome is influenced by the environment, with key ASVs assigning to common seawater taxa. Initial infection of chemoautotrophic symbionts appears to occur before settlement and metamorphosis in pediveligers of G. childressi, which were enriched for several chemoautotrophic taxa dominant in juveniles. The early acquisition of symbionts in pediveligers, previously assumed to occur after metamorphosis, may be a cue for settlement at a suitable seep habitat. In juveniles, the microbiome is composed of several methanotrophic symbionts known to inhabit G. childressi. Unexpectedly, we also detected sulfur-oxidizing symbionts in the SUP05 cluster and the heavy-oil degrading Cycloclasticus, which are not known to associate with this species. These findings challenged current assumptions and generated exciting questions regarding symbiont acquisition and colonization dynamics in G. childressi.

The influence of temperature on sexual phenotypic fate in winter flounder – A transcriptomic survey

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Many flatfish species show variations of sexual phenotypes as a function of thermal conditions during the first stages of development (Temperature-dependent Sex Determination"). With global warming, it is more important than ever to understand the mechanisms underlying the development of sexual phenotypes and the outcome on sex ratios. The objective of my project was to manipulate temperature conditions during the early life stages of winter flounder, a flatfish species found in the North Atlantic area to determine the thermo-sensitive life stage as well as mechanisms underlying modifications of sex. We produced different larval cohorts exposed to different temperature regimes from egg incubation to early juvenile stages and performed RNA-Seq surveys for each cohort at different periods of larval and juvenile development. We selected around 160 genes related to steroidogenesis and others that were previously identified as early sex markers in fish. Preliminary results indicate : 1) genes of interest are already expressed in early larval stages ; 2) the evolution of transcriptomic patterns differs among cohorts, but in the cohort exposed at higher temperature conditions differences among sampling times are much more differentiated ; 3) the expression of early female and male markers that vary among sampling periods and cohorts; and 4) an end-larval period thermo-sensitive stage with the most noticeable differences between cohorts after 40 days of development.

# Connectivity and larval dispersal of fish species with contrasting life histories in the deep-water region of the southern Gulf of Mexico

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Connectivity studies of the pelagic larval stage of fishes are crucial for comprehending their population dynamics and structure. Lagrangian trajectories from ocean circulation numerical models are a powerful tool for characterizing dispersal pathways of fish larvae and estimating the timing and spatial variation of spawning and recruitment. Forward tracking particle experiments were conducted to study the connectivity of five species with contrasting life histories that were sampled in the southern Gulf of Mexico's (GoM) deep-water region. The potential habitat of mesopelagic and neritic larvae was characterized by fitting generalized additive models to larval densities and ocean measurements, and used to delimit the origin of the particles. Two periods with contrasting mesoscale circulation were selected to evaluate particle retention or dispersion throughout the GoM: (1) an intense Loop Current anticyclonic eddy (LCE) interacting with the cyclonic eddy (CE) typically found in the Bay of Campeche, and (2) no interaction between a weak LCE and the CE. Larval transport was simulated with a high resolution assimilation model with hourly resolution (HYCOM + NCODA). A total of 1500 randomly distributed passive particles were seeded within the potential habitats at five depths within the first 200m and advected for 50 days. We hypothesized greater retention during lower mesoscale feature intensity and no interaction between the LCE and CE, and strong westward advection in the region of intense interaction between eddies. The results will provide insight on how larval habitat and mesoscale circulation influences dispersion and distribution, with implications for connectivity within the GoM.

## Baby Driver: Identifying likely drivers of redfish (Sebastes mentella) recruitment in the Gulf of St. Lawrence

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Overfishing and decades of poor recruitment caused Gulf of St. Lawrence (GSL) redfish, Sebastes mentella, populations to crash and stock biomass remained low until the mid-2010s. Unprecedentedly strong recruitment of the 2011 and 2013 cohorts have allowed redfish populations to rebound and talks of reopening the fishery are currently underway, but little is known about their early life ecology. In addition, redfish are categorized as a spasmodically-recruiting species, which produce strong recruitment cohorts on average once every 10 years. This characteristic makes it difficult to gather a large sample size of strong recruitment years in order to identify similar trends in environmental conditions over time using statistical models. The objective for this project was to identify likely environmental conditions that drive recruitment success of larval redfish using long term environmental datasets and a weight-of-evidence approach. We previously described early life trophodynamics and its relationship with growth, and used these results, to ultimately identify multiple likely oceanographic factors that play a role in redfish recruitment. We found a likely reliance of redfish recruitment on early phenology and high abundance of its key prey taxa, the copepod Calanus finmarchicus. We also proposed a relationship between the low volume of the GSL's cold intermediate layer and redfish recruitment, which we hypothesize increases larval survival immediately post-extrusion as larvae move up through the water layers towards surface waters. These results highlight the importance of investigating both physical and biological oceanographic conditions during early life stages in order to better understand recruitment variability.

#### Key to Reef Fish Conservation at the Texas State Aquarium

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The global marine aquarium supply is dependent on wild stocks of ornamental reef fishes. Overexploitation and harmful collection techniques targeting ornamental species threaten fish populations and reef ecosystems. Aquaculture can reduce demand on wild stocks while increasing our limited knowledge on the early life history of aquarium species. Standard protocols for egg collection and larval rearing in public aquaria can facilitate research and development towards large scale aquaculture by providing an untapped source of fishes from frequent voluntary spawning events. This research investigates the application of techniques to improve larval culture by characterizing morphological differences of eggs and larvae collected at the Texas State Aquarium (TSA). Samples collected from Living Coral Reef, a mixed species exhibit, were categorized by morphological features. Identities were validated using DNA barcoding at the mitochondrial Cytochrome C-Oxidase subunit 1 (CO1) gene region. Three species were confirmed with genetic barcoding: smallmouth grunt (Haemulon chrysargyreum), cottonwick grunt (Haemulon melanurum), and bicolor damselfish (Stegastes partitus). Differences in egg diameter were observed between H. chrysargyreum and H. melanurum. The larvae of S. partitus were recognized by characteristic pigmentation. Project data and resources were utilized to create an identification key for further aquarium application. Rapid sorting of mixed species can aid in assessing egg viability and documenting early development for implementing culture techniques of target species. This study discusses new applications in ornamental larval culture by strengthening current research, calling for further support of sustainable fisheries and commercial aquaculture, and presenting opportunities for outreach in the public sector.

## Experimental Methodologies for Examining Scope of Responses of Early Life-stages of Marine Fishes to Environmental Stressors

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The scope of biological response of an organism to environmental change varies and is a species characteristic. Some taxa show inherently greater levels of plasticity, have greater resiliency to environmental change, and display higher adaptive potential. Others are more conservative on all of these fronts. These patterns are of value in understanding both wild and captive populations. In order to achieve a more predictive understanding of how environmental change might affect marine life, science must progress from 1st-order, descriptive and qualitative assessments of effects, to more quantitative depictions of relationships between environmental drivers and biotic responses. Here we review the underlying principles of phenotypic variation then describe studies and experimental methods developed to reveal the scope of responses to environment drivers. Using examples from our work on recreational and commercial fish taxa, forage fish, status species, and laboratory models, we display patterns that would be unanticipated based on experimental designs with a small number of treatment levels. The approaches described have applicability to studies of climate change, ocean acidification, hypoxia, environmental toxicology, and aquaculture.

Advances in imaging technology and image analysis - applications in coastal systems

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In recent years, in situ imaging technology has evolved with considerable rapidity and can convey significant and novel insights into physical-biological patterns and processes. Optical methods, along with concurrent environmental sensors, can complement plankton net and acoustic sampling approaches. Combined with automated image analysis, such technologies have the potential to increase sampling efficiency thereby enabling either lower cost approaches, or expanded sampling leading to greater spatio-temporal resolution. Machine learning can reach broadly across these applications, as evidenced by its rapid rise across various disciplines and industries. The availability of machine learning allows for unique pattern/relationship seeking in model outputs, as well as for the rapid resolution of complex instrument outputs such as those from imaging systems or other observational sensors. Here we provide a brief review of the current state of imaging technology available to biological oceanographers, and some of the critical trade-offs. Included in the discussion is a view on the status of image processing, which has been a major constraint to extensive use of imaging technology. Recent work with the In Situ Ichthyoplankton Imaging System (ISIIS), and its associated automated image processing pipeline, is presented as an example of where progress has been made with imaging systems. Examples are given of the application of ISIIS from projects conducted within the California Current Ecosystem ranging from detailed perspectives of thin layer dynamics in Monterey Bay, California, to riverine influence on cross shelf plankton community distributions to utilizing near-real time analyses to drive adaptive sampling within hypoxic areas off the Oregon coast.

#### Forty years rearing marine fish larvae for aquaculture - are we there yet?

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Hubbs-SeaWorld Research Institute has been rearing various species of marine fish for aquaculture purposes since the early '80s. Current focal species are white seabass (Atractoscion nobilis), California yellowtail (Seriola dorsalis), and California halibut (Paralichthys californicus). These are three of 18 marine finfish species recently identified as "species of interest" in the USA by USDA and NOAA. Much has been learned during the evolution of mass production capabilities of these three native California species. At the broodstock stage, efforts have been focused on understanding and maximizing good egg and larval quality. Large marine brood fish present challenges in the amount of space and resources required for volitional spawning and conducting replicated manipulative trials is often impractical. Parentage analysis is helpful in understanding the breeding patterns of individuals in a group setting, including female-specific egg production and quality metrics. After the broodstock phase, larval stages require the most attention to maximize quantity and quality of fingerlings. The natural life history of a species can offer a good starting point for controlling environmental conditions and formulating feeds for different life stages. Recirculating aquaculture systems (RAS) provide the greatest level of environmental control and offer more robust biosecurity, but bacterial imbalances can be problematic for young larvae. Survival of A. nobilis, S. dorsalis, and P. Californicus are consistently greater than 20% from egg to fully metamorphosed juvenile at around 60 days post hatch. From a production standpoint, they are currently classified as "commercially" (A. nobilis and S. dorsalis), or "technologically" (P. californicus) ready.

Effects of exposure to elevated temperature and different food levels on the escape response and metabolism of early life stages of white seabream, Diplodus sargus

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Climate change is expected to limit food availability, due to increased ocean stratification, which will isolate the surface layers of nutrient-rich deep waters, potentially affecting primary production, with cascading effects to the entire trophic web. However, experimental studies addressing the effects of environmental stressors on behavioural and physiological responses of fish are frequently held under optimal feeding conditions, which might mask the real response of individuals to the stressor. Thus, addressing the interacting effects of food availability and other climate change related stressors on species responses assumes particular relevance, but, to date, has been poorly investigated. Here, we aimed at evaluating the effects of higher temperature and food availability, as individual and combined stressors, on the escape response and routine metabolic rate (RMR) of late-stage larvae of white seabream, Diplodus sargus. Temperature treatments were adjusted to ambient temperature (19°C), and a high temperature (22°C). Feeding treatments were established as high ration and low ration (50% of high ration). Escape response and RMR were measured after 4-weeks. Warming did not induce changes in behaviour or metabolism, but food shortage, interacting with high temperature, led to an increase in latency response and a significant reduction in RMR. The current study provides relevant experimental data on fishes' behavioural and physiological responses to the combined effects of multiple stressors. This knowledge can be incorporated in recruitment models, thereby contributing to fine-tuning of models required for fisheries management and species conservation.

## Momma's Larva: maternal investment and oceanographic conditions influence early growth and survival of rockfish

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Larval fish mortality is incredibly high, but small changes in mortality rates can lead to large changes in recruitment. Therefore, identifying factors that affect larval mortality is critical for understanding drivers of fish population dynamics. Recent work suggests maternal provisioning can dramatically affect the susceptibility of larvae to starvation and predation, which are major sources of larval mortality. We employed otolith microstructure analysis to examine patterns of maternal provisioning and how it influences larval traits of 8 rockfish species collected from 1998-2013 in California Cooperative Oceanic Fisheries Investigations (CalCOFI) surveys within the southern California Bight. Otolith nuclear radius, a proxy for maternal investment, was driven by the oceanographic conditions adult females experienced in the fall prior to winter larval release. In addition, core size was larger further from fishing ports possibly because these locations were historically less fished and contained older, larger females and/or had inherently better habitat quality than sites closer to shore. Otolith nuclear radii were significantly positively related to larval growth rate and age at time of collection, indicating that individuals with higher maternal investment grew faster and were more likely to survive early life stages. These results indicate that maternal investment is influenced by oceanographic conditions and potentially habitat characteristics and that the degree of maternal investment affects rockfish early survival.

Acclimation to low pH does not affect the thermal tolerance of Arbacia lixula progeny

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As the ocean warms, the thermal tolerance of marine invertebrates is key to determining their distributional change, where acclimation to low pH may impact the thermal range of optimal development. We compared thermal tolerance of progeny from a low pH-acclimated sea urchin (*Arbacia lixula*) population from the CO<sub>2</sub> vents of Ischia (Italy) and a nearby population living at ambient pH. The percentages of normally developing gastrulae and two-armed larvae were determined across 10 temperatures representing present and future temperature conditions (16–34°C). Vent-acclimated sea urchins showed a greater percentage of normal development at 24 h, with a larger optimal developmental temperature range than control sea urchins (12.3°C versus 5.4°C range, respectively). At 48 h, upper lethal temperatures for 50% survival with respect to ambient temperatures were similar between control (+6.8°C) and vent (+6.2°C) populations. Thus, acclimation to low pH did not impact the broad thermal tolerance of *A. lixula* progeny. With *A. lixula*'s barrens-forming abilities, its wide thermotolerance and its capacity to acclimate to low pH, this species will continue to be an important ecological engineer in Mediterranean macroalgal ecosystems in a changing ocean.

Plasticity of Larval Dispersal-Related Traits in Response to Changes in Parental Environment in a Coral Reef Fish

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Larval dispersal is an essential driver of marine fish metapopulation dynamics. Dispersal distance is highly variable among individuals within a species, however we currently do not know the causes of this variation. One likely cause of intraspecific variation in the pattern of dispersal is that there is dispersal plasticity. Many terrestrial studies have found plastic, adaptive dispersal responses to environmental cues, especially by offspring in response to their parental environment. Despite evidence across terrestrial plants, insects and vertebrates, dispersal plasticity has only been hypothesized in marine fishes. In this study, we test the hypothesis that parents in high-quality and low-quality environments will produce larvae that differ in dispersal-related traits using a lab population of *Amphiprion percula*. We measure swimming speed and body morphology of larval offspring in response to parental diet. We find a significant effect on larval body size, with larvae from low-quality treatments having greater body length, body depth, muscle area, and fin area, potentially facilitating greater dispersal distances. We found no significant effect of parental diet on critical swimming speed. These results may advance our understanding of the causes of variation in patterns of larval dispersal by potentially revealing an overlooked process of plasticity of dispersal-related traits.

## Larval Transcriptome Shows Transgenerational Effects of Parental Diet on Metabolism in a Marine Teleost

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Prior research on Red Drum (Sciaenops ocellatus) demonstrated that parental diet alters the fatty acid and lipid composition of offspring even when larvae are reared under uniform conditions, and this affects antipredator performance of larvae. Since parental diet controls yolk composition and yolk is the sole source of nutrition for embryos and early larvae, this finding indicates the presence of nutritional programming of offspring metabolism. The physiological pathways affected by this nutritional programming are not known. Three tanks of adult red drum were fed a diet of shrimp and then switched to a diet of sardines or herring, and multiple spawns were collected from each tank while on each diet. Larvae from these spawns were reared on a common diet until 21 days posthatching then sampled for differential gene expression analysis (TagSeq method) of whole larvae using the entire larval transcriptome (7,242 genes). There were 353 significantly differentially expressed genes between the parental diets. Biological processes related to ATP production (mitochondrion), protein synthesis (ribosome), and lipid metabolism (endoplasmic reticulum) were significantly enriched. Specifically, larvae from the parental shrimp diet had significantly higher expression of genes related to ATP production and protein synthesis and lower expression of genes related to lipid metabolism. This indicates that the shrimp diet fed to adults programmed an elevated metabolic rate in larvae. What are the implications for larval ecology? This programming response could increase the risk of starvation mortality in larvae when climatological or anthropogenic events substantially alter availability of certain prey types to adults.

Thermal tolerance of European sardine (Sardina pilchardus) larvae

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Sardine larvae survival is strongly dependent of temperature, and the inter-annual variability of temperature experienced by the larvae during the spawning season is strongly related to recruitment strength variability. Previous laboratory experiments studied the relation between sardine larvae vital rates and temperature during the first weeks after hatch, but there is currently no information on the thermal tolerance of sardine larvae throughout the ontogeny. Tolerance limits for sardine eggs and early larvae are very narrow, and high mortality are observed for larvae reared at temperatures above and below 17 and 13°C, respectively. Experiments were conducted to investigate how thermal tolerance evolve later in the larval stage. Sardines were reared at optimal (16°C) and higher temperatures (19 and 22°C) to test for effects on survival, growth, behaviour, swimming and oxidative stress parameters, throughout ontogeny. While no larvae survived at 22°C until 14 days post-hatching (dph), and survival was severely impaired at 19°C, tolerance to temperature increased sharply with age. At 25 dph, survival rates were similar between 16 and 19°C, while growth rate increased with temperature. From 15 to 33 dph, survival rate at 22°C was lower and growth was similar for larvae reared at colder temperatures, but the tolerance to higher temperatures clearly increased with larval age. These results show that larval dispersal models should parameterize larval temperature tolerance limits taking into account ontogeny and that the two first weeks after hatching are the most critical in terms of vulnerability to temperature.

Message in a Bottle: Archived DNA Reveals Marine Heatwave-Associated Shifts in Fish Assemblages

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Marine heatwaves can drive large-scale shifts in marine ecosystems but studying their impacts on whole species assemblages can be difficult. Here, we leverage the taxonomic breadth and resolution of DNA sequences derived from environmental DNA (eDNA) in the ethanol of a set of 23-year longitudinal ichthyoplankton samples, combining these with microscopy-derived ichthyoplankton identification to yield higher-resolution, species-specific quantitative abundance estimates of fish assemblages in the California Current Large Marine Ecosystem during and after the 2014–16 Pacific marine heatwave. This integrated dataset reveals patterns of tropicalization with increases in southern, mesopelagic species and associated declines in important temperate fisheries targets (e.g., North Pacific Hake (Merluccius productus) and Pacific Sardine (Sardinops sagax)). We observed novel assemblages of southern, mesopelagic fishes and temperate species (e.g., Northern Anchovy, Engraulis mordax) even after the return to average water temperatures. Our innovative preservative derived eDNA metabarcoding and quantitative modeling approaches open the door to reconstructing the historical dynamics of assemblages from modern and archived samples worldwide.

Maternal investment does not explain starvation resilience in marine larvae

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Egg size and maternal energy endowments are major themes in early life-history strategies. Here we report a series of experiments to mimic future changes in phenology of food availability and reproductive timing in a changing ocean. Utilization of lipid, protein, and carbohydrate energy reserves in sea urchin eggs and larvae (Strongylocentrotus purpuratus) were determined. The "Point of No Return" was calculated based on metabolic rate and utilization of egg energy reserves needed to support early life. Thermodynamic calculations reveal all egg energy reserves would be needed to support the first 6 days of development. Yet, compared to starting energy reserves in eggs, there was no change in protein or lipid content among larvae during the period of development tested (up to 12 days for unfed larvae). Starved larvae lived well beyond their calculated lifespan, maintained a constant metabolic rate, and a relatively high rate of protein turnover. A detailed series of transcriptomic analyses was undertaken to reveal possible mechanisms and source for the 'missing energy' used in development. Enhanced expression of a complex suite of amino acid transporter genes (known to transport dissolved organic substrates directly from seawater) was observed in unfed larvae, suggesting a molecular biological mechanism that accounts for the 'missing energy' observed during starvation trials. This evidence supports previous physiological studies of amino acid transport capacity in this species. These findings highlight the importance of the chemical environment, rather than a sole focus on egg energy reserves, when considering life-history strategies.

An index of spawning biomass of skipjack tuna (*Katsuwonus pelamis*) based on larval surveys in the Gulf of Mexico

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The development of fishery-independent indices of larval skipjack tuna, utilizing NOAA Fisheries Spring Ichthyoplankton Survey data collected from 1982 through 2019 in the Gulf of Mexico will be presented. Indices to evaluate trends in spawning stock biomass were developed using standardized data (i.e. abundance of 2-mm larvae under 100 m<sup>2</sup> of sea surface, sampled with bongo gear). Mortality rates, based on length distributions of collected specimens, are utilized to back-calculate larval numbers at 2 mm. The effects of an overall mortality rate versus an annual mortality rate on index model development were compared. Due to the large frequency of zero catches during ichthyoplankton surveys, indices of larval abundance were developed using a zero-inflated delta-lognormal model, including following covariates: time of day, time of month, geographic area sampled, sea surface temperature and year. The effect of the inclusion of an environmental variable (i.e. sea surface temperature) on index model development was evaluated. Finally, larval index trends were compared to those of spawning stock biomass produced in the stock assessment process, and the effects of inclusion of this index in the stock assessment model were evaluated. Work the body: Ocean acidification and links between feeding, conversion efficiency, growth, and mortality of larvae in a marine fish population

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For many marine species, exposure to ocean acidification conditions can increase mortality rates. This is especially true during the early larval phase when body sizes tend to be small and many larvae begin feeding on their own. Here we present evidence that mortality under OA conditions may be related to deficiencies obtaining food and converting that energy into growth and maintenance during development. In a series of laboratory experiments, larvae of California Grunion (Leuresthes tenuis) were reared under low and high pCO2 conditions (400 and 1050 µatm). We measured feeding rates, Specific Dynamic Action, growth, and mortality. Our results suggest that detrimental effects of ocean acidification are observable soon after the transition to external feeding (~3 days post hatch for California Grunion) and accumulate throughout the larval phase. Exposure to OA conditions resulted in a minor, but significant decrease in short term feeding rates. Perhaps more importantly, OA conditions had strong effects on Specific Dynamic Action – the summed energetic cost of food ingestion and biomass synthesis. For a standard meal size, energy devoted to SDA was ~33% lower for fish reared under OA conditions. This reduction in SDA was consistent with results of another set of experiments demonstrating reduced growth of fish developing under OA conditions. Finally, a comparison of growth and mortality that included replication at the level of full-sibling families suggested that families with low growth rates also had high mortality rates. Consistent with this general pattern, OA shifted average growth downward and mortality upward.

Physical transport and preferred prey effects on larval growth of Atlantic bluefin tuna (Thunnus thynnus) in the Gulf of Mexico spawning habitat

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Western Atlantic bluefin tuna (ABT) undertake long-distance migrations from rich feeding grounds in the North Atlantic to spawn in oligotrophic waters of the Gulf of Mexico (GoM). Previous research has shown that stock recruitment is strongly affected by interannual physical variability in the mesoscale features associated with ABT larval habitat, yet the nutrient sources and food-web structure of preferred habitat, the edges of anticyclonic loop eddies, are unknown. Here, we describe the goals, physical context, design and major findings of the BLOOFINZ-GoM project (Bluefin tuna Larvae in Oligotrophic Ocean Foodwebs, Investigation of Nutrients to Zooplankton). During the ABT spawning peak seasons in May 2017 and 2018, the oceanic GoM was surveyed for larvae, and and end-to-end process studies were conducted in five multi-day Lagrangian experiments (cycles) measuring hydrography and nutrients; plankton biomass and composition from bacteria to zooplankton; phytoplankton nutrient uptake, productivity, new production and growth rates; micro- and mesozooplankton grazing; particle export; and ABT larval feeding and growth rates. Study results highlight three significant findings: 1) advective transport from the productive shelf region is needed to resolve nitrogen budget and zooplankton production deficiencies in the oligotrophic offshore habitat of ABT larvae; 2) differences in availability of preferred prey (cladocerans) can alter larval growth rate trajectories during development, and 3) patches of abundant larvae are better explained as lateral transport from spawning sites along the outer continental slope region than by larval association with locally enhanced productivity along the outer edges of mesoscale eddies.

Biochemical strategies to manage food availability during the 'best and worst of times'

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A longstanding question in fisheries biology and aquaculture is the relationship between growth, recruitment, and food availability. Information remains scant, however, regarding a genetic basis of growth and survival within a population. Specifically, is there within-population variance among different larval families in their abilities to respond to fluctuating food environments? This question is relevant to production of "Blue Food," particularly in the context of global change. In this study, experimental crosses of pedigreed adults of the Pacific oyster (*Crassostrea gigas*) were used to generate a series of full-sibling larval families. These larvae were exposed to *ad libitum* algal food treatment ('best') and starvation ('worst'). A series of physiological and biochemical assays were used to assess the growth and starvation tolerance of each larval family. Surprisingly, the larval family exhibiting faster growth under *ad libitum* food was also the family with the best survival in the absence of food. The 'winning' larval family utilized more efficient biochemical strategies related to protein synthesis and turnover under both the 'best' and 'worst' food treatment conditions. These findings provide new insights into the genetic and physiological bases of adaptation to fluctuating food environments. Such information is of value to optimize yield in aquaculture to enhance food production.

# Modeling historical trends in larval stage duration of Lake Erie walleye to evaluate spring warming impacts on recruitment

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Climate variability can affect fish recruitment by altering early life growth and survival. To explore the potential impact of climate-driven changes in spring warming on recruitment, we used bioenergetics and hydrodynamic models to estimate the duration of the larval stage of Lake Erie walleye (*Sander vitreus*) during 2003-2014. Using historical environmental variability during this period, we sought to learn how anticipated spring warming might affect walleye recruitment by altering larval stage duration (LSD). We estimated LSD for six hatch dates during the spring under three levels of larval walleye consumption. Larval stage duration decreased with later hatch dates relative to earlier ones, owing to faster spring warming later in the spawning season. Larval stage duration in the east and west basins of Lake Erie were found to be largely unrelated, with significant declines in modeled LSD in the east basin of Lake Erie through time. Using our modeling results, we discuss how future, warmer conditions that result in shorter LSD might differentially affect the recruitment of local spawning populations of walleye across Lake Erie.

Sardine larvae dispersion and survival on the Iberian Current System: a numerical modeling experiment

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Management of small pelagic fish is confounded by the high interannual variability of recruitment strength. It is thought that environmental conditions experienced by the early life stages (eggs and larvae) are responsible for such variability. A set of different models were used to simulate the ocean conditions and the transport and survival of sardine early life stages in the Iberian Current System. They consist of a high-resolution simulation of the Iberian Current System (IBv2.0) with the hydrodynamic model ROMS, covering the entire Iberian Margin at a resolution of ~1.8km and 60 vertical levels, whose outputs are used as background for lagrangian simulations performed with the model Parcels coupled to an Individual Based Model of sardine eggs and larvae. The IBM for sardine eggs and larvae was developed based on previously published vital rates of the species in relation to key oceanographic factors; it was coupled to the Lagrangian model Parcels to compute dispersion patterns for particles deployed from the known spawning grounds. The IBM simulates the different early life stages of sardine (egg, yolk-sac larvae, first feeding larvae), and considers the effects of temperature and food availability on growth, survival, and development throughout ontogeny. Different years, characterized by similar sardine spawning biomass and contrasting recruitment strength were simulated to identify the impact of the environmental variables on egg and larvae survival and recruitment variability. The identification of key oceanographic processes responsible for recruitment success would enable an improved management of the species.

A synthesis of larval bluefin tuna growth dynamics from otolith microstructure in the Gulf of Mexico

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The western Atlantic bluefin tuna (ABT) stock mainly spawns in the Gulf of Mexico (GoM) in spatiotemporally restricted areas within a narrow range of environmental conditions. To better understand how the environment influences larval growth, concurrent biotic and abiotic parameters were obtained from multiple larval surveys in the GoM. Otolith derived growth and larval growth conditions were compared from various sampling efforts during peak spawning in 2014-2018. Key findings from this extensive effort include that for the first time, inter-annual growth rates are reported and were similar for larvae from 2015-2017. Then, despite pronounced larval associations with certain mesoscale features, daily growth trajectories showed no marked differences across water masses and features. In addition,  $\delta$ 15N larval values from a 2014 survey revealed that preflexion larval growth potential appears related to maternal  $\delta$ 15N signatures, likely due to maternally transmitted factors. Finally, investigations of two nursery habitats in 2017-2018 showed that larval growth was driven principally by food limitation and ingestion of actively selected copepod nauplii and cladocerans, underlying the importance of considering preferred prey when assessing habitat quality. We also observed that habitats differed in prey availability and resulting growth during larval stages differed pointing to an ontogenetic dimension to habitat quality. Overall, we summarize the current state of ABT larval ageing dynamics emphasizing the western stock, with some eastern spawning examples. Future studies should include environmental drivers of larval growth because they can potentially improve recruitment estimates by reducing the uncertainty of current models utilized in the management of ABT.

#### The cost of living for larvae in a changing ocean: Insights from integrating genetic and physiological analyses

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All small animals need to sustain very high rates of mass-specific metabolism (set by Metabolic Scaling Law). For larval forms, this intrinsic high demand for energy is further impacted by rapid global ocean change (e.g., rising temperature; food availability). Predicting the limits to which larval forms can respond to future change is a major challenge. There is always biological variance in response to environmental change. This variance is not simply experimental error -- there are complex genetic components involved that cannot be understood from experiments that only manipulate environmental variables using wild-type organisms. Biological response variance needs to be partitioned by genetic analysis, integrated with environmental and physiological processes. Experimentally, defining tipping points is especially difficult -- perhaps impossible -- given the near-infinite combinatorial interactions of biotic and abiotic processes in a changing ocean. Applying quantitative analysis to define the cost of living and bioenergetic modeling of homeostatic stability provides an integrative framework for assessing the capacity of larvae to respond to change. Our approach ("Ask the Organism") is based on: (1) measuring the limits of organismal-level resilience; (2) determining physiological capacity for tradeoff strategies in allocation of cellular energy (ATP) to respond to environmental stressors; (3) identifying biochemical and molecular biological markers of resilience; and (4) determining the genetic bases of adaptive responses (defined in 1-3 above). Using marine invertebrate larvae, specific examples will be presented to illustrate how adaptive response and resiliency could be identified (evolutionary "Winners") in a changing ocean.

The Use Of Indices Of Wind-Induced Microturbulence as Indicators of Pre-Recruit Survival of Yellowfin Tuna Thunnus albacares in the Eastern Pacific Ocean

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The Inter-American Tropical Tuna Commission (IATTC) conducts research on the reproductive biology and early life history of yellowfin tuna at the Achotines Laboratory, Republic of Panama. Yellowfin broodstock have been spawning at near-daily intervals since 1996, and the resulting eggs, larvae and early-juveniles are studied in experimental investigations to study the effects of environmental and biological factors on pre-recruit life stages. A 4-year study was conducted at the Achotines Laboratory from 1997 through 2000. The study was designed to estimate the optimal range of microturbulence for yellowfin larval survival during the first week of feeding in the laboratory. Yellowfin larvae exhibited a bell-shaped survival response to microturbulence, with intermediate levels of turbulence yielding the highest survival rates. Larval growth and gut fullness were also estimated. Turbulent dissipation rates were converted to equivalent wind speeds using a boundary-layer model for wind-induced microturbulence. The analysis was expanded to estimate optimal wind speeds for yellowfin larval survival in mixed-layer habitats in the eastern Pacific Ocean (EPO). A correlation analysis between optimal wind speeds and yellowfin recruitment in the EPO was conducted for selected 2º x 2º areas of the EPO from 1987-2007. Generally, positive correlations were found for areas south of 10º N latitude and east of 100° W longitude. Correlations became negative in areas more westward and northward. The positive correlations in the southeast areas of the EPO became even stronger and statistically significant when the analysis was temporally restricted to certain guarters of each year. The potential influence of wind-induced microturbulence on pre-recruit survival of yellowfin tuna will be discussed.

## Decoding comparable morphologies: Pigmentation validated for identifying southern California Paralabrax larvae

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The distribution and trends in larval fish abundance are often used to assess the status and trends of marine fish populations. However, for closely related species whose larvae are morphologically similar, and whose genetic identities may be degraded by formalin preservation, unraveling species-specific larval abundances from long-term monitoring efforts presents a challenge. We used statistical methods and the molecular identities of 107 ethanol-preserved specimens to construct and test a taxonomic key based on pigmentation patterns observed in three species of *Paralabrax* (family Serranidae) from southern California. Previously, larvae of these species were not thought to be reliably distinguishable based on morphology or pigmentation. However, when using pigmentation characters paired with molecular identities, a Random Forest Classifier provided a tool for structuring and refining a taxonomic key to distinguish species. Following calibration and key refinement, the probabilities of achieving accurate and precise species classifications using our taxonomic key were >96%, indicating that ventral and pectoral fin pigmentation patterns can discriminate *Paralabrax* larvae. Importantly, we can now leverage existing and future ichthyoplankton survey collections to assess species-specific trends in larval abundance, without the need for expensive and lab-intensive genetic analyses used with formalin-fixed specimens.

Defining a visual luminoxyscape to describe oxygen and irradiance constraints on larval habitat in a changing ocean

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Vertically migrating planktonic organisms are exposed to large changes in abiotic stressors over space and time; physiological limits to these conditions can constrain animal distributions. Vision requires both suitable oxygen and irradiance to function; this is especially true for animals with high-resolution vision. Previous research has shown vision to be sensitive to reduced oxygen, with severe visual impairment observed at oxygen conditions above traditional definitions of hypoxia. We propose the concept of a 'visual luminoxyscape' to describe the habitat bounded by oxygen and irradiance limits for vision in larvae of California market squid, Doryteuthis opalescens, two-spot octopus, Octopus bimaculatus, graceful rock crab, Metacarcinus gracilis, and tuna crab, Pleuroncodes planipes. We use the results of invivo experiments to calculate the lower depth limit of the visual luminoxyscape, called the visual luminoxyscape depth, which is the depth at which oxygen and irradiance would be sufficient for > 50% visual function and mildly-impaired visual behavior. Taking advantage of long-term ecological monitoring in southern California by the California Cooperative Oceanic Fisheries Investigations, we examined how the visual luminoxyscape depth changes over time and space for larvae of each species. We found that conditions that limit vision are currently present in larval habitat, and that oxygen is more limiting than the minimum irradiance needed for vision. Seasonal and inter-annual variability drives changes in the visual luminoxyscape depth, and deoxygenation has caused a shoaling of nearshore visual habitat at a rate of 1 - 3.8 m/y over the last two decades for larvae of all species.

## Changes in the coastal ichthyoplankton assemblage of a rapidly changing Gulf of Maine: The view from a southern Maine estuary

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Patterns and trends in larval fish assemblages present a potentially informative approach to quantifying ecosystem responses in a rapidly changing Gulf of Maine, which has experienced one of the fastest warming trends of any marine system on Earth. Here, we present results from a long-term (2009 to present), high-interval sampling of ichthyoplankton entering the Webhannet River Estuary (Wells, ME, USA). Sampling was conducted during late flood tide to assess the ingress of larval fish from nearby coastal waters. To date, the collection includes nearly 9,000 individuals representing 36 species, of which four, Tautogolabrus adsperus (cunner), Ammodytes americanus (sand lance), Clupea harengus (Atlantic herring), and Pseudopleuronectes americanus (winter flounder) dominate the assemblage (>80% of the individuals captured), and cleanly resolves seasonal variability in the coastal ichthyoplankton assemblage. The clearest responses to climate change and variability are found in the occurrence of rare (and new) species into the region, such as the detection of larval Centropristis striata (black sea bass) following anomalous warming in 2012, and shifts in the abundance of common species that consistent with plausible population and ecosystem responses to climate variability in the Gulf of Maine. These results demonstrate the value of long-term sampling at coastal locations for increasing our understanding of the variability in larval fish assemblages as an indicator of coastal ecosystem response to ongoing climate change.

Tracing exploitation of egg boons: An experimental study using fatty acid biomarkers

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Coordinated spawning of fishes and other marine animals releases millions of planktonic eggs into the environment, known as egg boons, creating nutritional resource pulses. Eggs are rich in essential fatty acids and may be an important lipid subsidy to egg consumers. Fatty acids (FAs) are widely used as qualitative markers to trace or confirm prey-predator relationships. Our aim was to validate the application of FA tracers of egg consumption in a variety of potential egg consumers and to confirm that the selected species may be fish egg consumers. We conducted feeding experiments with a selection of taxa including a planktivorous fish, a ctenophore, and a crustacean. We fed these animals a common diet (Otohime or Artemia) and simulated egg boons for half of them by supplementing the common diet with Red Drum (Sciaenops ocellatus) eggs for 3-6 consecutive days every 10 days for 1-3 months. Controls didn't receive eggs. In all three species, FA profiles of consumers fed eggs was significantly different from that of controls 24 h after the last feeding of eggs. Consumers took on FA characteristics of eggs. However, their FA profile was similar to that of controls within 2-10 days after the last eggfeeding event. Overall, results indicated that certain combinations of FAs could potentially be used as biomarkers of egg consumption within a short period of time. We conclude that FAs can be used as biomarkers of recent egg consumption in several marine species, validating their use for assessing exploitation of egg boons in nature.

Within-family variation in larval growth and survival is controlled by different genes

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Variance and covariance of larval growth and survival have generally been viewed in marine ecology and fisheries science as consequences of the environment. Genetic variation in these traits is less well understood, so we investigated the dependence of growth and survival on genotype in a full-sibling larval family of Pacific oysters (Crassostrea gigas). We serially screened sibling larvae, reared in the same (replicated) environment, to produce small, medium, and large size classes, which had necessarily had different growth rates. We genotyped parents and individual larvae sampled from each size class with a panel of mapped DNA markers, enabling identification of quantitative trait loci (QTL) contributing to variance in larval growth and survival. Larval mortality (86%), as expected, was essentially wholly determined by seven viability QTL, which mapped widely across the genome. Strikingly, a substantial portion of variance in growth among the size classes was determined by five QTL spread across three chromosomes; the combined effects of just two of these QTL accounted for almost 40% of measured variation in larval size. Little overlap of viability and growth QTL suggested that variation in these larval traits was controlled by different genes and that better survival of faster growing larvae in nature ("bigger-is-better") is likely not a genetically determined correlation. A large genetic component of variation in larval growth rate raises important questions about the role of such inherent variation in marine recruitment and the adaptation and evolution of larval forms.

# Abundance and condition of American lobster postlarvae in surface coastal convergences: patterns and potential processes

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The last stage in larval development of the American lobster (Homarus americanus), the postlarvae, occurs in surface waters, and previous studies suggest they may aggregate in surface convergences such as fronts. Surface convergences, i.e. surface habitats featuring convergent horizontal currents, may also aggregate other materials and organisms that could provide shelter and food for postlarvae and thus enhance their condition. We tested these hypotheses by conducting a series of cruises in the Gulf of Maine, where we sampled 15 potential convergences in summer 2021, and in each case, a paired offconvergence habitat. We measured postlarvae abundance and neuston community structure, surface hydrography, acoustic backscatter, and circulation. Laboratory experiments, lipid analyses and image analyses on postlarvae measured survivorship, condition, and color of postlarvae sampled on and off potential convergences. While the highest postlarval abundances were found in convergences, abundance patterns on and off potential convergences were not consistent. Laboratory analyses indicated no survivorship or lipid profile differences among convergence and non-convergence individuals. However, image analyses revealed coloration variability. Physical measurements indicate convergence heterogeneity, and community analyses reinforced these differences, showing substantial heterogeneity among potential convergences in community structure and surface material. Our results reinforce that small-scale heterogeneities are highly variable but critical in the ecology of meroplankton, including the pelagic and neustonic habitats where lobster postlarvae are abundant.

Preliminary study on distribution, abundance, composition and diversity of ichthyoplankton in the West Coast of Sri Lanka.

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Studies on ichthyoplankton in Sri Lankan waters are scarce, despite its importance in fish stock assessment and management. In 2018, an ichthyoplankton survey was conducted along Sri Lanka's west coast from March to May. A WP2 net was used for sampling. A total of 7154 fish eggs and 202 larval fish were obtained from the collected plankton samples. Among the obtained larval fish, 85 were yolk-sac larvae. 82 were successfully identified and classified into 16 families while the remaining 37 were unable to identify as they were damaged. There were no significant differences in abundances of fish eggs (P > 0.05) or fish larvae (P > 0.05) between vertical and horizontal hauls. Although larval fish from the family Clupeidae (28.05%) were the most dominant among the identified ones followed by family Engraulidae (25.61%), Mullidae (13.41%), Leiognathidae (7.32%) and Pomacanthidae (7.32%), the majority of larvae were yolk-sac larvae (41.67%). Temporally, the highest larval fish and egg abundances were observed in March while spatially, northern region adjacent to the Negombo lagoon. The west coast's northern region showed a relatively higher diversity of fish larvae than that of the Southern region. Because of the significant diversity and abundance of fish eggs and larvae found in this study, the west coast is most likely a spawning and nursery area for demersal and pelagic fish. The current study generates baseline data on the early stages of ecologically and commercially significant fish families.

# Connecting the nearshore: The fundamental role of thermal stratification for larval transport

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Larval transport in the nearshore plays a central role in larval dispersal and connectivity of shallow water species; however, few studies have resolved the relevant scales of larval transport and patterns of larval distribution in this region. With a greater focus on biophysical modeling, we often lose sight of the limitations of how we approach larval dispersal studies, particularly in the nearshore environment. Biological (larval behaviors) and physical factors (responses to environmental cues and hydrodynamic conditions) that influence dispersal and settlement dynamics are often poorly resolved. We will share insights into larval transport mechanisms learned from our work with Southern California intertidal barnacles.

How maternal investment and environmental conditions impact larval condition of Northern Anchovy: implications for small pelagic fishery management and recruitment success

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Coastal pelagic species (CPS) have frequent population boom and bust cycles that are heavily influenced by environmental changes, and a major focus of fisheries management research since the early 20th century has been to understand what causes population fluctuations in CPS. For Northern Anchovy (Engraulis mordax; anchovy), it was believed throughout the 20th century that anchovy experienced high year-class strength when the Pacific Decadal Oscillation was in its negative, cold temperature phase. However, a 2014-2016 marine heatwave challenged this assumption when anchovy rose to near record high numbers following California's highest 3-year period on record for sea surface temperatures. To determine which factors influence anchovy larval survival, we analyzed how environmental conditions (temperature, salinity, chlorophyll a and small plankton volume) and maternal investment (indexed by otolith core diameter) affected larval body condition of 3-6 week old larvae (body length, body depth, and otolith outer band width, an index of growth rate) between 2009 and 2019. Generalized additive models demonstrated that otolith core width was highly, significantly positively correlated with larval condition, small plankton volume was moderately, positively correlated and there was a strong random effect of collection location. Although data was limited, we also identified a suggestive relationship between larval condition and recruitment. Overall, we concluded that maternal investment is a very important factor impacting larval anchovy survival. For fisheries research going forward, the importance of maternal investment should be further emphasized, and future studies should further analyze the relationship between larval condition and recruitment success.

Invasive species effects on larval fish density, growth and distribution in southeast Lake Michigan

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Pelagic prey fish biomass in Lake Michigan has declined since the early 1980s coincident with declining nutrient loads, food web disruption by aquatic invasive species (AIS), and variable piscivore predation. We hypothesize that biomass declines of adults also may be related to AIS effects on fish larvae. We compared densities, distributions and growth of larval fish sampled at at nearshore, mid-depth and offshore sites before (from April-September in 1983 in SE Lake Michigan) and after (April – September at a nearby site in 2010, 2015, and 2021) invasions by the predaceous cladoceran Bythotrephes and Dreissena mussels. Densities of deepwater sculpin (DWS), bloater (BLT), yellow perch (YP) and rainbow smelt (RBS) larvae all were much higher in 1983, but alewife (ALE) larvae densities have remained unchanged amongst years. Depth-stratified sampling offshore indicated that in 1983, BLT and DWS larvae densities were highest in the neuston and epilimnion, but recently are most dense in the metalimnion. Zooplankton prey biomass also has declined since 1983 resulting in lower growth rates of BLT, ALE and YP larvae. Possible causes of changes in larvae density, growth rate, and vertical distribution offshore include: (1) exposure to UV radiation and predation risk under increased water clarity owing to Dreissena-mediated reductions in plankton biomass; (2) replacement of high quality zooplankton prey in larval diets by lower quality mussel veligers; and (3) decreased spawning populations of ALE and RBS. Decreased adult planktivorous fish facilitated Bythotrephes expansion and epilimnetic predation pressure, resulting in zoo- and ichthyoplankton redistribution to deeper waters.

Energy budget of stage-specific responses to hypoxia in a coastal fish

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Hypoxic zones are expanding and occurring more frequently in coastal oceans and estuaries, often coinciding with stressful temperature and pH levels. The physiological mechanisms and energetic costs of coping with hypoxia vary by life stage in fish. Dynamic Energy Budget (DEB) theory accounts for varying energetic inputs and demands at each life stage to explain whole life-cycle organismal effects of stressors and scale these effects up to higher levels of biological organization. We used data on an estuarine fish, the Atlantic silverside (Menidia menidia), to model the implications of early life responses to hypoxia for the full life-cycle energy budget. The data for the model came from the literature and a series of experiments in which embryos and larvae were reared in combinations of acidification and oxygen treatments, as well as two experiments in which embryonic and larval metabolic rates were measured as oxygen was gradually depleted. The objective of the modeling is to use DEB processes to explain the organismal effects of hypoxia observed in early life stages of M. menidia, including changes to metabolism, reduced hatch size and survival, and increased time to hatching. We modeled this using a simplified formulation of DEB theory (DEBkiss) with an added stress function to quantify energetic consequences across a range of oxygen levels. We hypothesized that the parameters for ingestion, assimilation efficiency, and maintenance rate could explain patterns in the data. This model will ultimately be expanded to incorporate previously documented interactions between hypoxia and acidification.

# Combined Effects of Ocean Acidification and Food Availability on Metabolism and Mortality of California Grunion Larvae (*Leuresthes tenuis*)

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Studies investigating the effects of ocean acidification (OA) on larval fish metabolism and mortality have produced varying results, suggesting other factors may have a moderating influence on responses to OA. To investigate how food availability may affect the responses of metabolic rate and mortality to OA, larvae of California Grunion (Leuresthes tenuis) were raised under two pCO<sub>2</sub> concentrations representing current day and a possible future climate change scenario (Ambient=406.8  $\pm$  13.7  $\mu$ atm & OA=1049.5  $\pm$ 67.3 μatm) and two feeding treatments (80 & 160 Artemia spp. Nauplii individual<sup>-1</sup> day<sup>-1</sup>). Metabolic rates were measured as routine oxygen consumption over a 20-minute period and analyzed using a linear mixed effects model. Mortality was measured by counting the number of dead larvae each day and analyzed using a generalized linear mixed effects model. For metabolic rate, there was a significant effect where low feeding levels decreased metabolic rates in comparison to high fed treatments (p=0.041, n=1909 from 49 families). In addition, the combination of low feeding levels and high CO<sub>2</sub> led to a further decrease in metabolic rate (p=0.019, n=1909 from 49 families). For mortality, there was a significant, interactive effect such that when food levels were low mortality rates were significantly elevated for fish in the high CO<sub>2</sub> treatments (p=0.0326, n=168 groups from 49 families). The synergistic effects of OA and food availability on both mortality and metabolic rate is important to consider as ongoing changes in seawater chemistry are likely to occur in concert with shifts in food web dynamics and prey availability.

# The effects of combined stressors, ocean warming and ocean acidification, on Pacific cod (Gadus macrocephalus) early life stages

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The North Pacific is simultaneously experiencing ocean warming (OW) and ocean acidification (OA), which may negatively impact the early life stages of important fish species. Pacific cod is an important commercial fisheries species with demonstrated sensitivity to OA and OW, but their combined impacts are unknown. Through a 5-week long experiment, we reared Pacific cod eggs and larvae at 3, 6, 10C and pH of 8 (ambient) and 7.3 in a factorial design. Overall for Pacific cod, temperature had a dominant effect on survival, growth and condition, low pH tended to increase growth and reduce mortality, and there were interactive effects of temperature and pH on some responses. Across both pH treatments, egg survival at hatch and larval survival at the end of the experiment was lowest at 10C, and the effect of pH varied across temperatures. Length and mass-specific growth increased with temperature; there was a stage-specific effect of pH on growth with faster growth at low pH at an early stage. There was a significant interactive effect of temperature and OA on larval condition (deviance in myotome height at a specific length). Fish in highest condition were observed at 6C at ambient pH, while fish in the 3C treatment were in the lowest condition with those in the ambient pH in lower condition than in low pH. These results show the complexity of stage- and trait-specific responses to co-occurring climate stressors, and the importance of investigating the effects of OW and OA in combination.

Are ephemeral fine-scale oceanographic features hotspots for ichthyoplankton? Testing the potential of eddies to entrain, sustain and retain larvae.

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Frontal eddies form on the shoreward edge of boundary currents, on average at weekly intervals and can last up to several weeks. This duration allows zooplankton production and completion of the larval stage of fish, but may be too ephemeral or dynamic for predator or competitor populations to detect or establish. Therefore, frontal eddies may provide a suitable offshore nursery habitat, by entraining and retaining inner-shelf water, preconditioned with coastal plankton and ichthyoplankton, into an upwelling favourable cyclonic eddy. In this seminar I briefly describe the behaviour of frontal eddies formed by three western boundary currents in the context of Bakun's "fundamental ocean triad", which incorporates three processes for the successful reproduction of fish: nutrient enrichment, food concentration and retention of larvae. I describe a pragmatic approach to quantifying entrainment of coastally spawned larvae in relation the ubiquitous larval myctophids. I conclude with how we can quantify the recruitment potential of larval cohorts (the larval Mortality/Growth ratio), without reading otoliths, based on their size frequency distribution. I present some preliminary findings from the CalCOFI archive on larval Pacific hake *Merluccius productus*, 1979 to 2021.

New Lens can Provide Novel Insight on Fish Recruitment

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Fisheries scientists have been attempting to resolve the mechanisms governing massive fluctuations in year class strength (recruitment) for more than a century. Elucidating processes affecting recruitment have proven difficult, but modern technologies are providing us with the potential to observe conditions at relevant scales and make significant progress towards understanding recruitment dynamics. In the California Current Ecosystem (CCE), we are currently applying multiple novel approaches to elucidate recruitment dynamics of northern anchovy (Engraulis mordax). Compound Specific Stable Isotope Analysis showed that the trophic position of larval anchovy prey predicted anchovy spawning stock biomass two years later, and that a sudden, lasting transition to higher trophic level prey in 1989 predicted a population collapse. Analysis of larval anchovy condition and otoliths also indicated that maternal effects and ocean conditions impact the quality of ~ 3-5 week old larvae, and that recruitment positively correlated with larval condition between 2009 and 2019. To provide insight on anchovy larval prey during low and high recruitment years, we are conducting Environmental DNA (eDNA) analysis of water samples and anchovy guts. To better resolve the role of oceanography on anchovy recruitment, we are running Individual Based Models that will trace the transport and survival of larvae from 2000-2020, a period of massive recruitment variability. In addition, we are using larval age and size frequency data to estimate rates of Growth and Mortality to evaluate recruitment potential from 1951-present. This holistic approach will hopefully shed light on the age old question of "what controls fish recruitment?"

Where are the fish? Larval fish sampling and transport modeling reveal vertical, spatial, and temporal distributions of Alewife in Lake Michigan

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The Cooperative Science and Monitoring Initiative (CSMI) is a binational effort to collect and synthesize data and to inform environmental management of the Laurentian Great Lakes. To elucidate fish recruitment bottlenecks, CSMI sampling in Lake Michigan has occurred once every 5 years across increasingly finer temporal and broader spatial scales. In 2010, 2015 and 2021, we sampled larval fish, plankton, water quality and optical properties monthly from nearshore to offshore in southeast Lake Michigan. We then related larval fish densities and distributions of Alewife Alosa pseudoharengus, a key prey fish, to variation in abiotic (temperature, PAR, UV for 2021) and biotic (chlorophyll A, zooplankton species-specific biomass) variables. Concurrent with sampling, we used a hydrodynamics model and larval fish back trajectory model to predict larval fish hatch locations and transport based on known ages, capture locations and dates of larvae. Across the 3 years, densities of Alewife larvae were primarily captured nearshore at the surface. Although initial larval densities were highest in 2015, a huge upwelling event in July advected larvae offshore, and reduced densities to below those observed in 2010, a year with no upwellings and much higher recruitment. In 2021, larval densities were lower than in 2010 or 2015, and this year was also characterized by a broad upwelling event. Use of the larval transport model, combined with high frequency sampling of water quality, plankton and larvae over space and time, has helped identify upwelling during the larval stage as a potential recruitment bottleneck.

# Productivity metrics of capelin (*Mallotus villosus*) spawning sites across habitats and bays in coastal Newfoundland, Canada

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In fish species with adherent eggs, spawning habitat characteristics are relevant for recruitment as they determine the conditions experienced in early life stages when mortality is highest. Capelin is a shortlived forage fish species that underwent a population collapse during the early 1990s in Newfoundland, Canada and has yet to recover. The aims of this study were to (1) compare temperature, timing of spawning and annual larval productivity in a bay monitored by the Department of Fisheries and Oceans (DFO) (Trinity Bay (TB)), and a more northern bay (Notre Dame Bay (NDB)) and (2) compare productivity metrics (i.e., temperature, egg density, percent dead eggs) between beach (intertidal) and subtidal habitats in NDB. The date of first spawning and annual larval productivity in TB and NDB were significantly positively related, suggesting that years with delayed spawning and low larval productivity are experienced shelf wide. This consistent variation in timing of spawning and larval productivity were not explained by temperatures at beach spawning sites as these did not vary consistently between the bays. Larval productivity was lower in NDB compared to TB. In NDB, larval densities at beach and subtidal spawning habitats were similar within the same year, but temperatures and the percent dead eggs were lower at subtidal sites. These productivity metrics, however, were not related to annual larval productivity. With the continued low productivity of Newfoundland capelin, understanding the contribution of specific habitats and bays to stock productivity is important to identify areas of high productivity for stock conservation.

Utilizing computer vision to count benthic organisms

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NOAA has been collecting millions of images of the seafloor using a towed camera system called Habcam. Annotating (marking) these images can be very labor intensive. As a consequence, only a small percentage of images are typically annotated, and then only for organisms that are of particular interest such as sea scallops (*Placopecten magellanicus*) and fish.

An alternative to human annotations is the use of computer vision/machine learning to automatically count benthic organisms. These automated annotators require a large amount of previously annotated training data to perform well. We developed a procedure to facilitate the training process. First, an automated annotator is trained using a relatively small number of manual annotations. Then the output from this automated annotator is checked and corrected using an R program. These corrected annotations are added to the training set and the model is retrained on the larger training set. This process is continued until a satisfactory annotator is produced. We used this iterative process to train automated annotators for sand dollars and sea stars. The use of automated annotations can increase the number of images and target organisms that are annotated, as well as reduce labor costs.

# Lunar and seasonal cycles in acorn barnacle *Chthamalus fissus* reproduction and settlement: Potential consequences for larval transport and dispersal

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The timing of life history events can have profound implications for populations and communities, especially those that influence species dispersal and recruitment. In benthic invertebrates, the majority of dispersal occurs during the larval phase of life. Yet the extent to which timing of reproduction (the beginning of the larval phase) directly influences timing of settlement (the end of the larval phase) is unclear, because biological and oceanographic processes can act to decouple these two events, and because reproduction and settlement are seldom studied together. Here, we used long-term (2014-2019) daily and weekly data from Bird Rock, La Jolla, California to look for lunar and seasonal cycles in reproduction and settlement timing of the acorn barnacle Chthamalus fissus, and we incorporate measurements of nearshore currents to examine the implications of reproduction timing on alongshore larval transport distances. We found evidence of clear lunar and seasonal cycles in reproduction, but only limited cyclicality on any timescale in settlement, possibly due to plasticity in larval development coupled with unpredictable onshore larval transport or dispersal. Even during periods of time when both series were collected concurrently, we found no cross-correlations between reproduction and settlement on any timescale. Our results indicate the complex nature of relationships between reproduction, larval transport, and settlement, which will be increasingly important to constrain in order to better document and predict species responses to climate change.

Oxygen supply capacity in larval fish

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Aerobic metabolism in larval fish has been measured with increasing frequency and precision in recent years, providing the opportunity for a greater understanding of the early development of oxygentransport systems in teleost fish. However, the wide variety of experimental, analytical, and reporting methods employed in these studies has obscured large-scale interspecific trends. Here, we summarize the available published data to assess critical oxygen levels (Pcrit) across species. The oxygen supply capacity method was used to standardize the methodology used to calculate Pcrit. Additionally, we report original data measuring these metrics in pre-metamorphic Common Snook. We found that in all observed species, larval fish do possess distinct Pcrit values, indicating active regulation of oxygen supply. Under ambient conditions, larval fish demonstrate higher Pcrit than adults of the same species. These trends were seen both in the representative respirometry data found in the published literature, as well as in our collected respirometry data. This suggests that typically, fish larvae have evolved to live closer to their thermal limits than older individuals. Thus, early life stages may be more vulnerable than juveniles or adults to increasing seawater temperatures at the warmer geographic edges of species ranges. Overwinter growth and energy allocation of Black Sea Bass juveniles in Long Island Sound

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Black Sea Bass (BSB, Centropristis striata), the northernmost serranid in the Northwest Atlantic Ocean, is a prominent example of climate change mediated range expansion in marine biota. Long Island Sound (LIS) has seen a particularly steep increase in BSB abundance of about 1,000% in the past decade, aided perhaps by exceptional LIS warming rates (4× global average) and reduced salinities that are generally conducive to BSB overwintering. We conducted two experiments on BSB juveniles: the first to quantify temperature-dependent growth effects of food availability (10-20°C) and the second to test how intermittent hypoxia exposure (0, 1, 3h at 2.5 mg/L DO) followed by a prolonged winter temperature exposure (6,12,18°C) would affect growth and energy allocation. Fish fed  $\geq$  2% of their body mass grew ~1-3x faster than those fed near-starvation in both mass and length, and allocated more lean mass to structural growth. Fish reared at 6°C grew 2x and 6× slower than those reared at 12 and 18°C, respectively, with no subsequent effects of hypoxia. However, fish initially exposed to 3h hypoxia accumulated 1.2 - 1.5x less fat mass than those in 0-1h hypoxia treatments regardless of temperature, suggesting that even brief hypoxic events in late-summer may alter BSB energetics and subsequent storage of fat reserves. Our results indicate that a warming LIS may soon become suitable overwinter habitat for this expanding meso-predator, portending further high recruitment and increasing abundance with inevitable food web consequences for LIS and other potential Northwest Atlantic coastal waters.

POSTER PRESENTATIONS

# The effect of presence and shape of naturally weatherted microplastic on Baltic spring spawning herring (*Clupea Harengus membras*) embryonic mortality

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Wide distribution and effects of microplastics in marine ecosystems is alarming. Microplastics are expected to increase the potential health risks to humans through food chain, but also limit the production of commercially harvested fishes. To date the vast majority of microplastics studies has focused on food web or toxicity effects whereas mechanical disturbance due to microplastics remains almost not known. Here, we studied the effect of physical disturbance of microplastics on the Baltic spring spawning herring (Clupea harengus membras) embryos. Our experiment involved three treatments: 1) no microplastic, 2) microplastic with sharp edge, 3) microplastic with rounded edge. We used the partially degraded microplastics particles of a garbage patch to reproduce the characteristics and the diversity of the microplastics that organisms encounter in their natural environments. Baltic herring was collected from commercial fishermen trapnets and transported to the laboratory where eggs of survived individuals were fertilized. The fertilized herring embryos were deployed in 0.75 L bottles at natural environmental conditions (water temperature 10°C and salinity 3.5 ‰). Mortality was checked in 6th day after eggs fertilization. Dead embryos were counted in each bottle. The results suggest that the presence and shape of microplastics affected survival of herring embryos, although differences between treatments were only marginally significant at p = 0.10. Mortality was lowest without microplastics (2.5%), intermediate with sharp microplastics (4%) and highest with smooth microplastics (6.5%). We plan to conduct similar experiments for the autumn spawning Baltic herring this year (in September).

Trophic ecology and connectivity of Bay anchovy (Achoa mitchilli) in Chesapeake Bay tributaries

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Bay anchovy Anchoa mitchilli is a dominant forage species in the Chesapeake Bay, and our goal was to evaluate the effect of abiotic and biotic habitat conditions on its young-of-the-year trophic ecology. We coupled historical data with contemporaneous measurements of abundance and stable isotope composition of larval (n = 180) and juvenile (n = 180) bay anchovy. Bay anchovy specimens were collected in horizontal tows from surface and mid-water (60-cm diam. net, 280-µm mesh) monthly from June to September of 2018 from the Patuxent and Choptank river tributaries and in August 2018 from the Chesapeake Bay near the Patuxent River mouth. We compared the diet, trophic position, and trophic niche of YOY bay anchovy between these two nursery tributaries and evaluated correlations between trophic metrics and body condition metrics. We also examined the effect of riverine discharge on the role of tributaries as foraging grounds (i.e., is upstream trophic connectivity increased under higher discharge conditions). We found that: 1) larvae in the upper rivers in June had equilibrated to tributary food webs, 2) isotope data suggest recruitment in July, inferred from larvae appearing with mainstem trophic signatures, 3) newly recruited larvae and juveniles started to equilibrate to the tributary food web, suggesting local residence, feeding and growth, 4) all anchovy caught in the rivers show the same temporal patterns, regardless of where in the river they were caught, and 5) larvae and juveniles showed the same temporal patterns, suggesting no major ontogenetic shifts between life stages.

Distribution of meroplankton above methane seeps in the Gulf of Mexico and Western Atlantic Margin

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Cold seeps on the Western Atlantic Margin (WAM) and in the Gulf of Mexico (GOM) are chemosynthetically productive ecosystems populated by shared species of chemosymbiotic Bathymodiolin mussels that host dense communities of endemic invertebrates. Presence of shared adult species suggests connectedness yet similarities in larval assemblages and potential dispersal between sites remains unclear. To explore larval distributions, we examined meroplanktonic density and diversity above these cold seeps. We collected demersal meroplankton above cold seeps at one GOM site in Fall 2021 and 8 sites in Spring 2021 (3 WAM and 5 GOM seeps) using the newly developed SyPRID plankton sampler on the AUV Sentry. We also collected demersal meroplankton at the Florida Keys, the boundary between the GOM and WAM, and in Spring 2021 we collected samples at the thermocline above all sites. On average we sampled >2,000m3 water per sample at ~700m3 per hour. We recovered an average of 0.0207 larvae per m3 and 168 larval morphotypes from 14 phyla. The diversity of meroplankton represented both cold-seep and non-seep benthic invertebrates with only weak similarities between community assemblages of each tested depth per site. Nine morphotypes were found in both seep regions, each spanning at least two depth classifications, with five of these morphotypes present at the Florida Keys boundary. One shared morphotype was a larval Bathymodiolin mussel and was found at demersal depths of most sites, and the thermocline at one site. Additional genetic analysis will help decipher the origin of the remaining targeted morphotypes.

Mortality and Clonal Maintenance of Crown of Thorns (Acanthaster sp) Larval Populations

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For marine invertebrates that have a dispersive larval stage, the peril of the plankton is considered to be a major mortality bottleneck. Echinoderms that reproduce asexually through larval cloning have a mechanism to replenish populations through binary fission and budding with the fragmented bodies having the potential to regenerate to form a new complete larva. We followed populations of the bipinnaria larvae of the crown of thorns seastar (*Acanthaster* sp, COTS) through time to determine natural mortality rates and the influence of cloning as a compensatory mechanism to help maintain the number of individuals. The types of clones and mechanism of body fragmentation was also characterised. In populations generated from multiple different populations, the incidence of cloning varied, where parental source influenced cloning ability. This inherent plasticity and ability of COTS to amplify their population numbers will have important consequences with respect to the dynamics of COTS in the plankton. Salinity Tolerance of the Bipinnaria Larvae of Acanthaster sp. (Crown-of-Thorns Starfish)

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Crown-of-thorns starfish (COTS) outbreaks cause widespread coral mortality across the Great Barrier Reef. When COTS larvae are in the plankton, nutrient rich coastal runoff simultaneously generates phytoplankton blooms, which potentially enhances their food levels and success to precipitate an outbreak, and lowers salinity. The potentially deleterious effects of decreased salinity on COTS larvae has not been considered. The impact of decreased salinity and increased food on COTS bipinnaria larvae was investigated. The percentages of larval normal morphology and larval survival were determined in response to 7 salinities and 3 algal food density treatments, at four time points. Salinity performance curves showed that salinity was the major factor determining larval performance. At 24 h the optimal salinity ( $S_{opt}$ ) ( $\geq$  90 % larval survival) was ~26–34 ‰ and the salinity with 50 % mortality ( $LS_{50}$ ) was 21.62 ‰. By 96 days the LS<sub>50</sub> had increased to 24.57 ‰, showing a narrowing of salinity tolerance over time. Salinities for the sharp onset of deleterious effects approximated ~22–25 ‰. These are levels that larvae would experience in a runoff plume. Counter to the paradigm of enhanced larval success, these findings indicate a negative effect of runoff for COTS larvae. Comparison of sampling efficacy of bongo and manta plankton nets in Southern California

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Ichthyoplankton sampling is used worldwide to track spawning dynamics of myriad fishes. Since the late 1970s the California Cooperative Oceanic Fisheries Investigation (CalCOFI) program has consistently utilized both oblique bongo and neustonic manta net tows to quantify abundances of larval fishes. Although many publications have analyzed bongo data to track fish assemblages, mantas have received much less attention, and the utility of this net to sample species relative to bongos is currently unclear. Here we evaluate the capacity of bongo versus manta nets to quantify abundances of fish larvae in southern California from 1978-2015. First, we examine what species are most abundant in the manta versus bongo samples. Second, we assess whether there are any significant correlations between larval fish abundances in bongo nets compared to manta nets. Third, we evaluate impacts of day versus night sampling for manta versus bongo nets. Although many larval fish species were abundant in both types of nets, some species were much more abundant in mantas than bongos (e.g., California Grunion, Pacific Saury) and vice versa (e.g. rockfishes, sanddabs, and mesopelagic species). In addition, whereas abundances were highly correlated between nets for some species, there are no correlations for others. Finally, there was no systematic bias for day versus night sampling for either net. We conclude that although manta data are rarely used to track species abundances, mantas are better suited for quantifying abundance of several common species in southern California whose larvae are neustonic.

# Effects of ocean acidification and warming on fertilization, egg survival and size at hatch of early life stages Polar cod (*Boreogadus saida*)

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Warming temperatures and rising pCO2 levels are co-occurring and present the need for a greater understanding of multistressor effects on marine species. Polar cod (Boreogadus saida), a pelagic, keystone species, serves as a high energy food source for marine mammals and seabirds. Early life stages of Polar cod are sensitive to temperatures above 3.5°C but effects of elevated pCO2 are unknown. We examined fertilization, hatching success, and size at hatch of Polar cod in two experiments. Polar cod eggs obtained from a captive broodstock were fertilized at pH 7.6 and 8.0 with fertilization success in 3 replicates scored after 24 hours incubation at 1°C. Fertilization success varied across egg batches (obtained from different females), but was higher at pH 8.0 than 7.6 in 12 of the 15 batches. In a separate experiment, mixed parentage groups of eggs were incubated from 5 and 8 days post fertilization (dpf) to hatch under a factorial design of 3 pH (7.5 or 7.7 or 8.0) and 2 temperature (1°C or 3.5°C) treatments. High temperature reduced survival and time to hatch and resulted in higher rates of malformations. Higher temperatures also resulted in larvae hatching at a smaller size (length) but with increased yolk area. Elevated pCO2 appeared to have no effect on hatching success, size at hatch or yolk area. These observations of elevated pCO2 levels affecting fertilization success, but not egg development, demonstrate stage-specific effects and the need to consider climate variation at all life stages of marine fishes.

Construction of a Quatrefoil Larval Fish Light Trap Modified for Low-cost, Easy Reproduction

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The use of light traps is a widely accepted method for the passive collection of larval fishes. This presentation describes modifications to the quatrefoil light trap design and the materials used in its fabrication – resulting in significant improvements in economy of build, ease of construction, and compact disassembled profile for easy transportation. Constructed from readily available materials using common tools, this trap has been developed to be built without access to a workshop. The design and construction of this trap are described in detail for unrestricted use with attribution.

Dominance and widespread distribution of Southern bluefin tuna (*Thunnus maccoyii*) among the larval tuna assemblages in the eastern Indian Ocean

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Southern bluefin tuna (Thunnus maccoyii, SBT) is a valuable, highly migratory species widely distributed across temperate oceans of the southern hemisphere. Mature adults migrate to a single known spawning site between Java and Australia in the eastern Indian Ocean (IO), but information about SBT early life history is largely based on surveys conducted > 35 years ago. As part of the second International IO Expedition, the BLOOFINZ-IO (Bluefin Larvae in Oligotrophic Ocean Foodwebs, Investigations of Nutrients to Zooplankton) Program sampled the SBT spawning grounds in 2022 during the peak spawning period (January-February). Larval sampling was complemented by multi-day Lagrangian experiments to assess mesoscale variability, nutrient sources, primary productivity, foodweb structure, trophic fluxes and their influences on SBT larval trophodynamics and growth. SBT larvae were broadly prevalent in the warm (26°C), low salinity (~34 psu) mixed layer of the Argo Abyssal Plain in waters characterized by strong density stratification, low eddy kinetic energy and low chlorophyll a concentration. Our shipboard identifications showed SBT larvae dominating (~86%) the Scombridae abundances throughout the study area. In addition, a subset of presumed SBT eggs and larvae were genetically confirmed by sequencing a fragment of the mitochondrial cytochrome C oxidase subunit I gene. Our preliminary findings reaffirm active and daily SBT spawning events in the region. As part of this international collaborative effort, the recently funded INDITUN (MICINN-Spain) project will seek to explain trophodynamics and growth potential advantages of SBT larvae relative to other co-occuring species across habitat quality gradients of the spawning region.

Physiological responses to a warming ocean: Phenotypic contrasts among larval families

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The capacity to which organisms can tolerate rising temperature is a major concern in global change biology. Although it is well established that metabolic rate increases with temperature, less is known about the extent to which standing genetic variation within a species could confer physiological resilience to higher temperature for an evolving natural population. To assay for such variation, we conducted a series of genetic crosses of adult broodstock (Pacific oyster, Crassostrea gigas) to produce 21 different larval families (pedigrees were confirmed with mapped DNA markers). Measurements at different temperatures were used to quantify the physiology of thermal sensitivity among larval families throughout development. Thermal sensitivity was consistent within a larval family during growth, but showed significant differences among families. We identified specific larval families that under conditions of rising temperature: (1) increased cellular energy (ATP) supply through respiration and, conversely, (2) decreased ATP demand to support protein synthesis. The consequences of these differential thermal sensitivities for energy supply and demand are larval phenotypes with increased biosynthetic efficiency at higher temperature and larvae with a larger ATP pool that could be allocated to cope with additional environmental stressors. Further testing of transgenerational ("egg-to-egg") inheritance of these traits could contribute to a greater understanding and prediction of physiological resilience under scenarios of future climate change.

Increasing spawning ground knowledge for marine fishes through metabarcoding

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Characterization of fish egg assemblages contributes to the identification of spawning grounds. This information is useful for biodiversity assessments, ecological early life history studies, and fisheries management. However, species-level identification based on the egg morphological characters is challenging or impossible. Molecular identification using genetic markers (barcoding) like cytochrome oxidase I (COI) has improved the taxonomic resolution achieved for individual eggs. We implement high-throughput sequencing of barcodes (metabarcoding) for fish egg assemblages collected throughout the Gulf of Mexico during the GOMECC-3 cruise in the summer of 2017 with oblique bongo tows with 335 um nets. Stations included shallow (ca. 20 m) shelf stations as well as the deep-water region (bottom depth > 1000 m). A ~313 bp COI amplicon was used in 19 fish eggs-sorted samples containing 4,771 eggs in sum. Collectively, 108 species were identified, corresponding to 82 genera and 43 families. We analyze the diversity, species composition, community structure, occurrence, and the relationship between species presence/absence and sea surface temperature and salinity. Overall, we show how metabarcoding can be successfully applied to broad spatial scale studies of natural marine fish egg assemblages leading to expanding our knowledge on marine fish spawning.

Genetic and Morphological Identification of Formalin Fixed Larval Fishes: How Long Is Too Long?

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Identification of larvae of fish is usually based on assembling a developmental series of wild-collected larvae, usually formalin preserved, using pigmentation patterns, morphology and fin meristics. However, for many species, larvae are still undescribed, or there are only limited descriptions of larvae development. Formalin fixation of larval fish was previously thought to prevent genetic sequencing compared to ethanol-preserved larvae. In this poster, we detail the results of an integrative taxonomic approach based on morphology, imaging and DNA barcoding of the mitochondrial (mtDNA) cytochrome c oxidase subunit (COI) gene. We used this approach in both cultured yellowtail kingfish, Seriola lalandi and wild-sourced fish larvae fixed in 5% formalin. DNA barcoding and genetic species identification were 100% successful in S. lalandi fixed in formalin for up to 6 months, while barcoding of wild-caught fish larvae enabled species identification of 93% for up to 8-week formalin-fixed specimens. While COI genetic identifications from the in-field experiments were patchier than the controlled experiments, our study highlights the possibility of re-covering suitable DNA from formalin-fixed larvae for up to six months. This was achieved by applying DNA extraction methods that use de-cross-linking steps and species identification based on both full-length reference and mini-barcodes. Our study provides a practical framework for undertaking both morphological and genetic identification to document the larval development of previously undescribed species from historic and current formalin-fixed samples collected around southern Australia.

Environmental effects on Iberian sardine larvae distribution, condition and growth

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The Iberian sardine (Sardina pilchardus, Walbaum 1792) has a high socioeconomic value being the main target of the Portuguese purse-seine fishery. Recent years have shown clear signs of a full recovery of this fishery resource, after a decade of reduced abundance due to a prolonged period of low recruitments. The effect of environmental variables on larvae distribution, nutritional condition and growth rates were examined based on samples collected during the autumn of 2017 and spring of 2018, years that produced rather low Iberian sardine recruitments. Larvae were less abundant at the beginning of spawning season, when they were mainly restricted to the more productive and colder water masses. In spring, larval stages presented a more homogeneous distribution, with higher densities on the west coast, where ocean temperature was particularly cold that year. Generalized Additive Models showed a significant influence of spatial and temporal variables on the larvae density and depicted a negative influence of temperature. Larval condition was significantly higher in autumn and was the lowest in spring on the west Portuguese coast. The larvae condition revealed to be mostly influenced by intrinsic variables (e.g. length), and the effects of each of the environmental variables tested were non-significant. On the other hand, temperature and chlorophyll-a showed a significant positive effect on larvae growth rates. Results seem to indicate that, although sardine larvae have a preferred habitat in cold productive waters, these two environmental variables have a strong influence on survival potentially acting as main drivers of the recruitment success.

Diatom-specific polyunsaturated aldehydes negatively affect larval zebrafish (Danio rerio)

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Diatoms are ubiquitous in marine planktonic and benthic environments and are common diets for many lower-trophic organisms. Certain species of diatoms produce polyunsaturated aldehydes (PUAs) that can exist in particulate and dissolved forms. Diatom PUAs are known for negatively affecting the fecundity of their primary consumers, including invertebrate grazers like copepods and echinoderms. However, little is known about the effects of diatom PUAs on vertebrates that are sympatric with diatom populations, and that may be exposed to dissolved PUAs. The purpose of this study was to test whether dissolved diatom PUAs affect the early life stages of a model vertebrate, *Danio rerio* (zebrafish). To test for this, mixtures of the diatom PUAs decadienal, heptadienal, and octadienal were used in three developmental stress experiments. In one experiment, embryos 24 hours post fertilization (hpf) were exposed to 3 concentrations of PUA mixtures for 6 days. At 7 days post fertilization, a significant reduction in overall larval size was observed under the two highest PUA treatments. In a second experiment, embryos exposed at 24 hpf showed a significant reduction in heart rate after 2 days of PUA treatment. In a final experiment, embryos exposed to PUAs at 2 hpf showed lower survival compared to controls. This study reveals the potential for PUAs to affect vertebrates that co-occur with diatoms and highlights the need for future studies using ecologically important vertebrates associated with PUA-producing diatoms.

## Larval fish ingress and vertical distribution in the Aransas Pass Inlet system (TX, USA)

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In the Coastal Bend region of Texas several proposals for desalination plants are currently underway. These plants would take in water from the adjacent bay and coastal inlet and dispose of the brine using a diffuser creating a plume of higher salinity and temperature in the coastal inlet. The proposed developments have caused concern that especially vulnerable larval fish and crustaceans would be harmed when passing through the inlet from their Gulf of Mexico spawning grounds to their estuarine nursery habitats both in the public and scientific communities. Changing vertical position is a key behavior of larvae during selective tidal stream transport, which is considered one of the two main mechanisms to pass a coastal inlet together with passive transport. By sampling the Aransas Pass Inlet and its three tributaries, in the vicinity of one of the proposed desalination plans, during day and nighttime and during both incoming and outgoing tides at three different depth strata we aimed to improve our knowledge of the vertical position of larval fish and crustaceans during their passage through the channel. Preliminary data analysis revealed that larval densities occurred throughout the whole water column but were greater in the deepest stratum and nighttime hours. There may be an adverse effect on the larvae's development and survival depending on the larvae's sensitivity to abrupt changes in salinity and temperature which differs by species and developmental stage.

# Combined method study expands knowledge of larval habitat usage of shallow bottom fish Aprion virescens

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Although Aprion virescens (green jobfish, locally uku) is a popular fisheries species both commercially and recreationally in the main Hawaiian Islands (MHI), little is known about their early life history. To better define Essential Fish Habitat (EFH) for this snapper species and advance an ecosystem-based management approach, we investigated the physical characteristics of ocean areas used by larvae and pelagic juvenile uku. A comprehensive literature review and Indo-Pacific wide specimen inventory was combined with an examination of archived ichthyoplankton samples from the Island of Oahu. Our final dataset consisted of fewer than 300 records of individual larval uku collected throughout MHI from 1967 to2012. We characterize size and habitat patterns for 230 larvae from preflexion (3 mm) to pelagic juveniles (30 mm total length) with associated environmental data (temperature, salinity, seasonality and depth). Larval observations from archived samples (2-6.5 mm total length) were restricted to the top 30 m of the water column and were typically found 1--27 km from shore. Larvae from the literature (3-30 mm total length) were found 0.5-88 km from shore. We also evaluated sizes of settled benthic juveniles and discovered a knowledge gap; juvenile uku between 30 and 90 mm in length have yet to be recorded anywhere in the Pacific, and thus their habitat use during this critical settlement period is unknown. Our results extend existing information about larval uku habitat and highlight the need for more targeted studies on the early life stages of this commercially and culturally important species.

Diet and feeding niche of larval rockfishes (Sebastes spp.) in the Southern California Bight

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Survival through the larval phase greatly affects the population dynamics of most fishes, and a major factor affecting larval growth and survival is the capacity to feed on preferred prey. Here, we examine the feeding ecology of *Sebastes* spp. larval rockfishes in the Southern California Bight (SCB). Rockfishes of the family Sebastidae comprise a highly diverse and abundant family of fishes in the SCB and are both economically and ecologically important. Larval rockfishes were collected in a series of Bongo net tows throughout the SCB in fall 2020 and winter and spring 2021. Their stomach contents were extracted, identified, and enumerated. Larval diets were found to shift between succeeding developmental stages of copepods as a function of larval ontogeny, changing from Calanoid and Cyclopoid nauplii in preflexion larvae to predominantly Calanoid copepodites in the postflexion stage. Clear differences in relative contributions of important prey items were also observed spatially, particularly for early development stages. Larvae generally became more specialized feeders as they developed, but the degree of specialization observed between growth stages varied even across small spatial scales. These findings provide evidence of preferential but changing feeding during larval Sebastidae ontogeny, and draw attention to the importance of examining fine-scale spatial variation when evaluating conditional factors for larval recruitment success.