



the ALC LARVAL FISH AND CONFERENCE

CONFERENCE PROGRAM AND ABSTRACTS



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PICES



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Scientific Committee

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Welcome Message

On behalf of the Early Life History Section of the American Fisheries Society, we welcome you to the 46th Annual Larval Fish Conference, here in the beautiful Portuguese capital of Lisbon! We have both a stunning venue (the Gulbenkian Foundation) and a truly outstanding scientific and social program. With over 90 registered delegates (from 23 countries!), 58 speakers and 26 posters, the 2023 Conference promises to be a success!

No matter whether you are attending this Conference for the tenth time or are here for the very first time, we hope you experience this week as every bit as amazing and inspiring as we always have at every LFC we have attended!

So please enjoy and make the most of the 46LFC!

Ana Faria and Susana Garrido



General Meeting Information

Internet

Wireless internet (Wi-Fi) will be available free of charge at the main venue. Join the FCG Free WiFi.

Registration

You can register on Sunday 7th, from 17h00-20h00, and from 8-11th registration will be opened during the whole day.

Name badges

Participants are required to wear their name badge to all sessions and social events. Entrance into sessions is restricted to registered participants only.

Oral presentations

With the exception of the Plenary Sessions, oral presentations are scheduled for 20 minutes (15 minutes for presentation + 5 minutes for questions). Presenters must prepare their talks in either PowerPoint (.ppt or .pptx) or PDF (.pdf) format. Please ensure that unusual fonts, and any videos or sound files are embedded in the PowerPoint file (or transferred onto the presentation laptop). To save time between presentations, presenters will not be permitted to use their own laptop. Please, therefore, make sure that your talk gets uploaded to the conference laptop prior to your session.

Please name your talk using the code in the program (under «CODE_AUTHOR». E.g., «S3:Garrido»).

Student talks will be concentrated into sessions from Monday to Tuesday to allow the judges time to make their decisions prior to the awards reception on Wednesday evening.

Poster presentations

The poster session will be held Tuesday evening. The poster boards will be available all-day Tuesday. The stands for the posters will be dismantled on Thursday morning. Please place your poster after the first coffee break on



Tuesday in the board displaying the number assigned to your poster (see program). Posters must be no larger than 1189 x 841 mm in overall size (A0 format). We will provide mounting tape to place posters.

Student Travel Awards

The ELHS offers a number of student travel awards. There are three awards given by the ELHS: the Sally Leonard Richardson Award for the best student paper presented at the Larval Fish Conference, the John H.S. Blaxter Award for the best student poster at the Larval Fish Conference, and the Elbert H. Ahlstrom Lifetime Achievement Award. The ELHS also awards Grace Klein-MacPhee Student Travel Grants to facilitate graduate student participation at the Larval Fish Conference. Best student awards will be announced during the conference banquet, on Wednesday 10th.

Early Career Event

The Early Career Committee hosts a workshop on “Scientific communication to a general audience”, on Tuesday 9th. Conference attendees are invited to participate in an engaging, activity-based workshop focused on communicating findings from primary literature and personal research to a general audience. This event was developed by the Early Life History Section Early Career Committee and will be hosted by Dr. Marta Moyano (NIVA, Norway). While this workshop is tailored toward early career researchers, all are welcome.

Larval Fish Identification Workshop

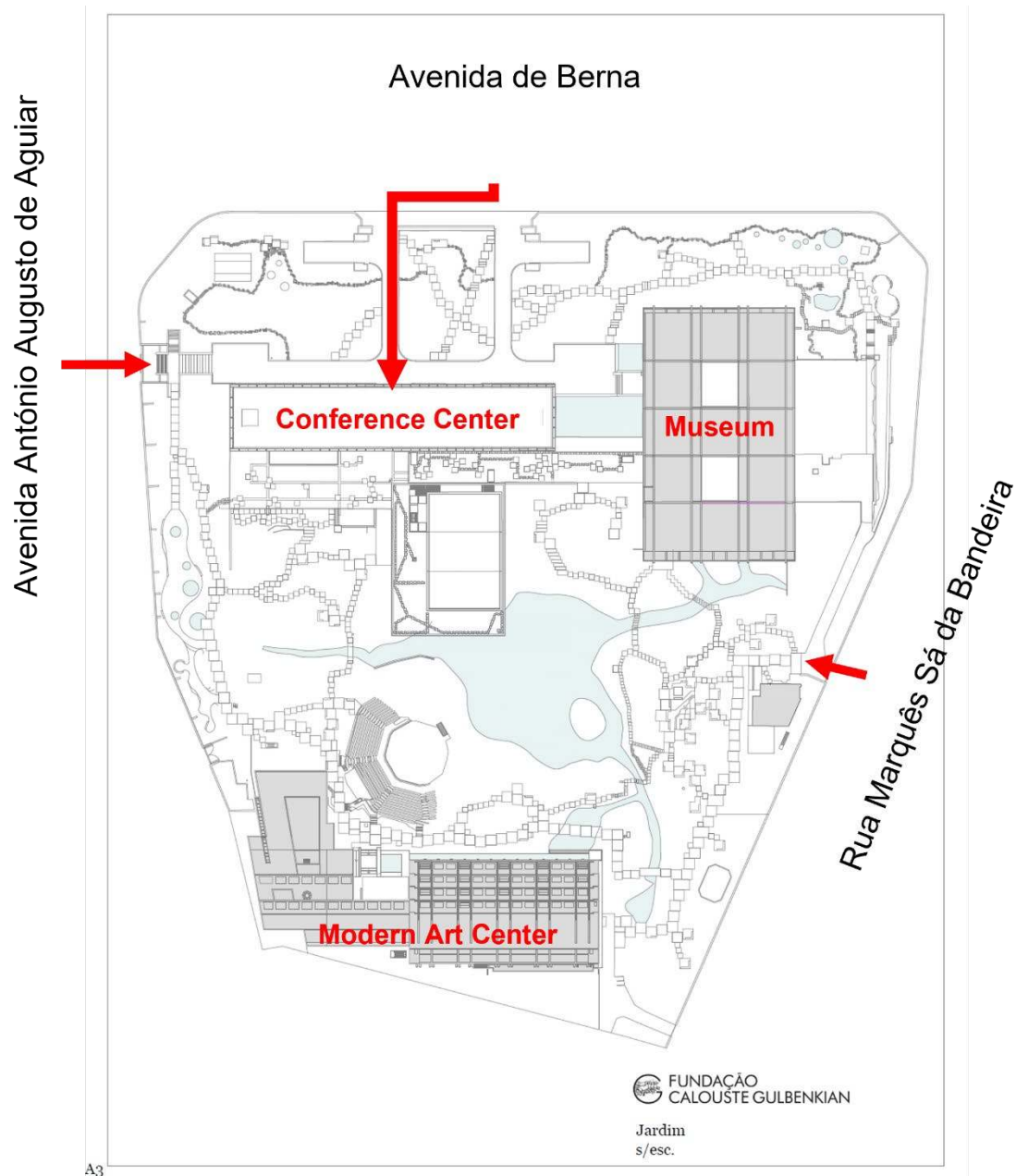
A Larval Fish Identification Workshop will be held on Monday 8th, hosted by Dr. Nalani Schnell (MHNH, France) and Professor Pedro Ré (University of Lisbon, Portugal). Participation will be limited to 15 people, with students having priority. Please tick on the corresponding box at the registration page if you are attending!

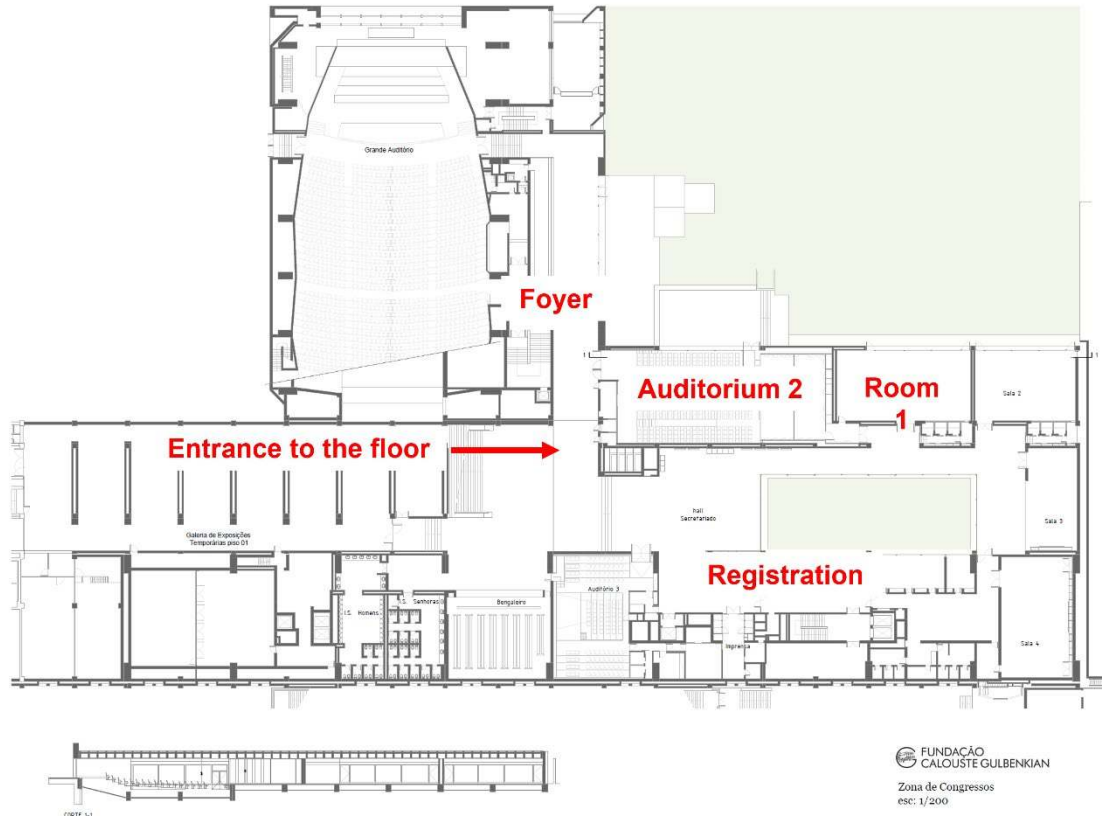
For any enquiry, please contact: lfc46@larvalfishconference.com



Venue

The conference venue, located at the headquarters of the Gulbenkian Foundation, boasts a privileged location in central Lisbon, right next to the famous Gulbenkian Museum, and the Center for Modern Art, with its surrounding beautiful gardens. Gulbenkian Foundation is located in Av. de Berna, 45A, 1067-001, Lisbon.







Theme sessions

S1. Global change effects on early life stages.

Convenors: Ana Faria, Filipe Ribeiro and Hannes Baumann

Early life stages are expected to be most vulnerable to changes occurring in aquatic environments, but the effects are likely to differ across species, developmental stages (embryos, larvae, juvenile) and habitats (freshwaters, estuaries and ocean waters). In this session we invite presentations that examine direct or indirect effects of key environmental factors that are changing as a consequence of climate change and have been defined as determining factors for the maintenance of physiological functions in fish. These include temperature (both gradual warming and extreme events, such as marine heatwaves), ocean acidification and hypoxia. We also encourage contributions about research on emerging contaminants, microplastics, noise, light pollution, habitat alteration and parasites. We particularly value submissions that focus on the combined effects of these stressors and on the ability of individuals to acclimatize to changing conditions.

S2. Recent trends in marine larviculture.

Convenors: Núria Baylina and Raquel Quirós-Pozo

Knowledge of larval biology of marine teleost is still on early stages for many species, particular for those targeted by the marine ornamental trade and for commercial aquaculture diversification. Also, there is a limited number of species that have successfully bred, both for biological research and/or commercial aquaculture purposes. On this regard larval ontogeny and zootechnics are still poorly studied for the majority of marine teleost species. This lack of basic knowledge is a big challenge not only for larval commercial production (food and aquarium trade), but also for fisheries stock management. More recently, overfishing, climate change and the increase in threats to natural hatcheries such as mangroves, estuaries or coral reefs habitats destruction, marine fishes breeding has gained increased interest and importance not only for aquaculture species diversification but as a powerful tool for marine species conservation purposes. The aim of this session is to include studies that focus on larval development description and identification, including studies addressing ontogeny, prey selectivity, feeding behaviour, nutrition, but also zootechnical description and optimisation of the larval rearing systems, amongst others.



S3. Larval Trophic Ecology.

Convenors: Susana Garrido, Pedro Ré and Marta Moyano

The importance of food for larval survival has motivated a large number of studies on the feeding ecology of fish larvae. These studies involve a large number of techniques such as studying the diet composition of field caught larvae through prey visual identification, or less frequently using fatty acid biomarkers and more recently using molecular techniques. Also, trophic position and trophic niche breadth were studied by analysing the stable isotopic composition. Feeding experiments in laboratory-controlled conditions allowed to obtain several parameters such as ingestion rates, prey requirements, feeding behaviour and relation of feeding rates with other environmental variables such as temperature. In this session we welcome studies focusing on the feeding ecology of fish larvae from marine and freshwater environments, including those focusing on ontogenetic shifts of diet composition, taxonomic differences, diet broadness, and spatial and temporal variability of feeding.

S4. Integrating and modelling early life-history processes and connectivity.

Convenors: Ana Machado and Stefan Koeningsstein

Early life history processes are influenced by the physical and ecological environment, and are important drivers of fish population and community dynamics. Physiological and ecological process rates quantified by empirical approaches are set within the spatio-temporal dynamics of the environment, which needs to be considered when assessing the overall impacts on a species' life cycle and productivity. New ecological modeling approaches are contributing to understanding fish early life history stages and the processes that lead to their variation in space and time. This session invites studies from ocean and fresh water systems which use numerical and conceptual models, to link empirical results to impacts on species and ecosystems. Approaches include individual-based models, biophysical larval dispersal models, energy budget models, recruitment, population and ecosystem models. We strive to better connect empiricists with modelers, and advance understanding of the sensitivity and response of fish populations to environmental variation and climate change.

S5. Larval survival and recruitment variability.

Convenors: Alexandra Teodósio and Vânia Baptista

The causes of recruitment variability are a key question in the ecology of marine communities, with consequences for global human population directly depending on marine resources. Several hypotheses have been proposed to explain recruitment of fish species, focusing on survival and growth of fish larvae, as well as the physical and



complex food web processes that sustain fish larvae in nursery areas. It is also known that fish larvae use sensorial and swimming abilities to detect and ingress into nursery habitats. In this session we welcome studies from all seas and inland waters focusing on the specific relevance of factors potentially determining recruitment success of fish larvae from marine and freshwater ecosystems, including survival and growth rates, swimming and personality, as well as their ontogenetic changes, spatial and temporal variability, habitat uses, and larval ecophysiological condition. Contributions to a deep understanding of recruitment variability from tropical, temperate or extreme aquatic environments are needed and will lead to an adequate management of marine ecosystems and key fisheries with direct consequences to an healthy ocean and quality of life increase.

S6. Application of “omics” in larval research.

Convenors: Laura Ribeiro and Paulo Gavaia

The application of analytical technologies has contributed to deepen the knowledge on how fish larvae cope with external challenges and successfully develop and reach juvenile stages. During the period where development, differentiation and maturation of tissues and important biological functions are taking place, integrated analytical approaches like histological, biochemical, enzymological, and molecular techniques, have contributed to understand the synchronism between structure and function at different developmental stages. Still analytical limitations exist related with small larval size, sampling conditions, among others. The recent application of “omics” (metabarcoding, proteomics, lipidomics, metabolomics, genomics and transcriptomics) to larval research have contributed in overcoming some of the technical limitations. In this session we welcome studies focusing on the application of omics technologies and other analytical tools, applied on studies with fish larvae from marine and freshwater environments, to gain deeper understanding of fish larvae biology, such as predatory capacity and prey selection, characterization of functional responses when exposed to different conditions, epigenetic effects, identification of molecules involved in tissue differentiation and regulatory pathways during development.

S7. Other Theme

Any other contribution of high quality not fitting the schedule will be included here.



PLENARY SESSIONS

How do the temperate pelagic fish larvae recruit to a suitable habitat (i.e. find a home)?

Maria Alexandra Teodósio

Centre of Marine Sciences, Universidade do Algarve, Faro, Portugal. E-mail: mchichar@ualg.pt

A series of complementary hypotheses have been proposed to explain the recruitment of marine and temperate pelagic fish larvae originated from pelagic eggs in coastal environments. In this talk, we present a complementary hypothesis describing the biophysical processes intervening in the recruitment of temperate fish larvae into estuaries. This hypothesis, the Sense Acuity And Behavioral (SAAB) hypothesis, recognizes that recruitment is unlikely if the larvae drift passively with the water currents, and that successful recruitment requires the sense acuity of temperate fish larvae and their behavioral response to the estuarine cues present in coastal areas. We propose that temperate fish larvae use a hierarchy of sensory cues (odor, sound, visual and geomagnetic cues) to detect estuarine nursery areas and to aid during navigation towards these areas.

Direct and indirect effects of ocean acidification on herring (*Clupea harengus*) and cod (*Gadus morhua*) larvae – a comparative approach

Catriona Clemmesen

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Rising atmospheric CO₂ levels and resulting changes in ocean pH (ocean acidification) have been shown to significantly affect the early life stages of fish with consequences relating to their recruitment potential. Results from laboratory and large-scale mesocosm experiments, where fish eggs and larvae are kept at different levels of CO₂, representing future climate change scenarios, are evaluated focusing on growth, energy allocation, survival, behavioral responses, otolith and bone formation, tissue damages as well as gene expression analyses. Using large-scale mesocosm experiments indirect effects relating to changes in the food web were analyzed and indicated potential compensatory mechanisms due to increased food availability. Results from parental acclimation experiments and from a latitudinal approach, comparing the same species from different sampling locations, showed adaptation potentials to CO₂ stressors. Overall, the analyses of the responses of herring and cod larvae, commercially and ecologically very important species in marine, temperate regions, showed some species-specific responses. Additionally, the environment experienced by the parents highly influenced the response of the fish larvae to the CO₂ stressor. By using modelling approaches to upscale the experimental results, the consequences of increased CO₂ levels on the fish population are evaluated.



The Shape of Things to Come: Resiliency of Marine Fish Early Life-Stages in a Changing World

R. Christopher **Chambers**

NOAA, Northeast Fisheries Science Center, Highlands, New Jersey USA, 07732. E-mail: chris.chambers@noaa.gov

Knowledge of how early life-stages of marine fishes respond to environmental variation is fundamental to understanding population recruitment, adaptive potential of individuals, and the likelihood of persistence of a species in a changing world. Our planet continues to be subjected to ever increasing levels of impacts from human activities with the extent and severity of biological responses expected to increase. Nursery habitats used by many marine fishes are under a myriad of environmental threats associated with these activities including changes in thermal, oxygen, and CO² regimes, and the introduction of persistent industrial contaminants. Revealing biological response functions to these challenges provide fisheries ecologists a means to glimpse into future recruitment and to identify optimal conditions for production in an aquaculture context. This presentation provides an overview of the principles of phenotypic responses to environmental challenges, the experimental approaches to revealing the occurrences and patterns of these effects, and the types of biological responses that might be expected in species subjected to these challenges.

Dispersal-relevant behaviour of fish larvae – measuring it, putting it into models and using a dispersal model incorporating behaviour

Jeff **Leis**

Institute for Marine and Antarctic Studies, University of Tasmania, Hobart 7001, TAS, Australia; Australian Museum Research Institution, William Street, Sydney 2001, NSW, Australia. E-mail: jeffrey.leis@utas.edu.au

Since the mid 1990s “the simplifying assumption” that marine fish larvae could validly be treated as passive particles whose dispersal was totally dependent on currents has been gradually discredited by increased understanding of the behavioural capabilities of the larvae. This has led to a generally accepted view that dispersal of marine fish larvae is a biophysical process dependent on both currents and larval behaviour. Yet, many contemporary dispersal models still fail to incorporate behaviour of larvae. This talk provides a historical perspective on larval-fish behavioural research over the past 30 years, and how behavioural data can be obtained and incorporated into dispersal models. Including the ontogeny of behaviour is essential as is incorporating behavioural variation at both within and among individual levels. Examples of questions that can be addressed with a validated dispersal model incorporating behaviour will be provided.



How Did We Get So Far From Where We Began? Looking Back on Our Team's Research on Maternal Effects

Lee A. Fuiman

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Research projects usually end with new questions. When those questions are pursued in subsequent projects and the cycle repeats, we can find ourselves far from where we began. More than a decade ago, we were investigating the causes of variation in performance of survival skills in red drum (*Sciaenops ocellatus*) larvae. We found evidence for effects of the quality (not quantity) of maternal investment in eggs on larval performance. What causes such variation in investment? What are the effects of varying maternal investment on larval performance? Can the effects be compensated? Which physiological pathways in larvae mediate the maternal effects? Do any of these findings apply to other species? What are the ecological consequences? Are there practical applications? This presentation will provide an overview of one team's research on maternal effects and how we went from predator-prey interactions and recruitment mechanisms to lipid physiology, nutrition, metabolic programming, transcriptomics, food-web ecology, climate, aquaculture, and stock-enhancement.

The importance of prey selectivity for revealing relationships linking prey availability to larval feeding success and survival potential

Dominique Robert

Research Chair in Fisheries Ecology, Institut des sciences de la mer de Rimouski, Université du Québec à Rimouski, Canada. E-mail: dominique_robert@uqar.ca Canada.

More than 100 years ago, Johan Hjort (1914) proposed the Critical Period hypothesis, which stated that strong year classes emerged when first-feeding larvae encountered large numbers of planktonic prey. Over the past decades, fishery scientists have tested the link between recruitment strength and plankton abundance, but very few studies have reported significant relationships. In this keynote address, I argue that the link between plankton abundance and larval survival remains concealed by the lack of knowledge of larval prey preference. While larval fish have long been considered generalist feeders, recent studies have shown that the vast majority of larval fish species are strongly selective from the first-feeding stage, and that a limited number of prey taxa are targeted by multiple larval fish species. I conclude that species-specific knowledge of prey preference is crucial to define the effective prey field and reveal relationships among zooplankton abundance, larval survival potential, and recruitment variability.



CONFERENCE SCHEDULE

Sunday, 7th May		Monday, 8th May		Tuesday, 9th May		Wednesday, 10th May		Thursday, 11th May	
		8h00-16h00	Registration desk available	8h00-16h00	Registration desk available	8h00-16h00	Registration desk available	8h00-16h00	Registration desk available
		9h00-9h15	Opening Ceremony	9h00-9h35	Plenary, Teodósio A.	9h00-9h35	Plenary, Fuiman L.	9h00-9h35	Plenary, Robert D.
		9h15-9h50	Plenary, Chambers C.	9h35-9h55	S6. Kolker M. *	9h35-9h55	S4. Catalan I.	9h35-9h55	S5. Sirois P.
		9h50-10h10	S3. Nasraoui S. *	9h55-10h15	S6. Ferreira A. *	9h55-10h15	S4. Berenshtein I.	9h55-10h15	S5. Paris C.
		10h10-10h30	S3. Avila L. *	10h15-10h35	S7. Marmelo I. *	10h15-10h35	S4. Machado A.	10h15-10h35	S5. Primo A.L.
		10h30-10h50	S3. Germain E. *	10h35-10h55	S7. Ranjula S. *	10h35-10h55	S3. Garrido S.	10h35-10h55	S7. Cumplido N.
		10h50-11h10	Coffee Break	10h55-11h15	Coffee Break	10h55-11h15	Coffee Break	10h55-11h15	Coffee Break
		11h10-11h30	S1. Siegfried E. *	11h15-11h35	S5. Guerreiro M.A. *	11h15-11h35	S3. Kanaya A.	11h15-11h35	S1. Réalis-Doyelle E.
		11h30-11h50	S1. Olaiola M.G. *	11h35-11h55	S5. Baptista V.	11h35-11h55	S3. Fiksen Ø.	11h35-11h55	S1. Cruz J.
		11h50-12h10	S1. Weisberg S. *	11h55-12h15	S5. Rutherford E.	11h55-12h15	S3. Moyano M.	11h55-12h15	S1. Muller C.
		12h10-12h30	S1. Fanny-Laure T. *	12h15-12h35	S2. Quirós-Pozo R.	12h15-12h35	S3. Murphy H.	12h15-12h35	S7. Miskiewicz T.
		12h30-12h50	S1. Magalhães S. *	12h35-12h55	S2. Schmidt N. *	12h35-12h55	S3. Fonseca P.	12h35-12h55	S7. Stern N.
		12h50-14h10	Lunch Break	12h55-14h15	Lunch Break	12h55-14h15	Lunch Break	12h55-14h15	Lunch Break
		14h10-14h45	Plenary, Clemmesen C.	14:15-14:50	Plenary, Leis J.	14:15-14:35	S5. Robert D.	14:15-14:35	S1. Rodrigues D.
		14h45-15h05	S4. Koenigstein S.	14h50-15h10	S2. Barraca C.	14h35-14h55	S5. dos Santos Schmidt T.	14h35-14h55	S1. Lima A.
		15h05-15h25	S4. Frøysa H.G.	15h10-15h30	S2. Castanho S.	14h55-15h15	S5. van Damme C.	14h55-15h15	S7. Thompson A.
		15h25-15h45	S4. Mason E.T. *	15h30-15h50	S2. Ozl. *	15h01-15h35	S5. Ferreira A.S.	15h15-15h45	Closing remarks
		15h45-16h05	S4. Nakita T. *	15h50-16h10	S2. Diogo P.	15h35-15h55	S1. Teletchea F.		
		16h05-16h25	Coffee Break	16h10-16h30	Coffee Break	16h00-18h30	Visit Oceanario de Lisboa		
		16h25-19h25	Workshop Larval ID	16h30-18h00	Early Career Event	18h30-22h00	Social Dinner		
				16h30-18h00	ELHS Business meeting				
				18h00-18h20	Tribute to John Bleaxter				
				18h20-21h00	Poster Session				
17h00-20h00	Registration desk available								
18h00-21h00	Ice-breaker at the Venue								



DAILY SCHEDULE

Monday, 8th May				
	SESSION/AUTHOR	CONVENERS	TITLE	
8h00-16h00	Registration desk available			
9h00-9h15	Opening Ceremony			
9h15-9h50	Plenary, Chambers C.	Faria A. + Garrido S.	The Shape of Things to Come: Resiliency of Marine Fish Early Life-Stages in a Changing World	
9h50-10h10	S3. Nasraoui S. *	Garrido S. + Moyano M. + Ré P.	Relationship between diet and morphology during larval development in different fish species of the Gulf of St. Lawrence	
10h10-10h30	S3. Avila L. *	Garrido S. + Moyano M. + Ré P.	Ecological implications of a cryptic species complex on the diet of early life stages of rainbow smelt in the maximum turbidity zone of the St. Lawrence Estuary	
10h30-10h50	S3. Germain E. *	Garrido S. + Moyano M. + Ré P.	Diet composition and prey selectivity of larval Winter Flounder in the southern Gulf of St. Lawrence	
10h50-11h10	Coffee Break			
11h10-11h30	S1. Siegfried E. *	Baumann H. + Faria A. + Ribeiro F.	Legacy effects of variation in larval size at hatch: A case study of California Grunion	
11h30-11h50	S1. Olaizola M.G. *	Baumann H. + Faria A. + Ribeiro F.	Will climate change impact the Baltic cod?	
11h50-12h10	S1. Weisberg S. *	Baumann H. + Faria A. + Ribeiro F.	Variability in fish phenology over 20 years of rapid warming along the Northeast US Continental Shelf	
12h10-12h30	S1. Fanny-Laure T. *	Baumann H. + Faria A. + Ribeiro F.	Co-exposure of tire wear particule coupled with increased temperature during the early life stages of a cold stenothermic lake fish, the char	
12h30-12h50	S1. Magalhães S. *	Baumann H. + Faria A. + Ribeiro F.	How is the plastic pollution situation in the Douro Estuary? Monitorization of microplastics and larval fish assemblages in a Portuguese urbanized estuary	
12h50-14h10	Lunch Break			
14h10 - 14h45	Plenary, Clemmesen C.	Faria A. + Garrido S.	Direct and indirect effects of ocean acidification on herring (<i>Clupea harengus</i>) and cod (<i>Gadus morhua</i>) larvae – a comparative approach	
14h45-15h05	S4. Koenigstein S.	Koenigstein S. + Machado A.	Projecting the unpredictable: Integration of early life stage processes to quantify recruitment and forecast fish stock dynamics	
15h05-15h25	S4. Frøysa H.G.	Koenigstein S. + Machado A.	Using a numerical model to propose a new laboratory oil exposure design	
15h25-15h45	S4. Mason E.T. *	Koenigstein S. + Machado A.	A Fish Tale As Old As Time: Larval Abundance Is Related To Spawning Stock Size - Or Is It?	
15h45-16h05	S4. Nakisa T. *	Koenigstein S. + Machado A.	Online Model Library of Larval Fish: A Tool to Better Understand Larval Survival in Marine Ecosystems	
16h05-16h25	Coffee Break			
16h25-19h25	Workshop Larval ID (Room 1)			

Tuesday, 9th May				
	SESSION/AUTHOR	CONVENERS	TITLE	
8h00-16h00	Registration desk available			
9h00-9h35	Plenary, Teodósio A.	Faria A. + Garrido S.	How do the temperate pelagic fish larvae recruit to a suitable habitat (i.e. find a home)?	
9h35-9h55	S6. Kolker M. *	Gavaia P. + Ribeiro L.	Quantitative species-level characterization of a larval community reveals narrow thermal affinity of invasive larvae, manifested across temporal and spatial scales	
9h55-10h15	S6. Ferreira A. *	Gavaia P. + Ribeiro L.	A proof-of-concept study to use DNA metabarcoding as a tool for species identification in ichthyoplankton samples	
10h15-10h35	S7. Marmelo I. *	Gavaia P. + Ribeiro L.	The use of macroalgae as an eco-innovative strategy to improve the immunity and antioxidant responses of juvenile farmed fish	
10h35-10h55	S7. Ranjula S. *	Gavaia P. + Ribeiro L.	Larval fish assemblage in Western coastal waters of Sri Lanka: Seasonal and spatial structure	
10h55-11h15	Coffee Break			
11h15-11h35	S5. Guerreiro M.A. *	Baptista V. + Teodósio A.	Ichthyoplankton communities at the northern Canary Current Upwelling Ecosystem - interannual variability and environmental forcing	
11h35-11h55	S5. Baptista V.	Baptista V. + Teodósio A.	What do we know about the little fish of São Tomé and Príncipe?	
11h55-12h15	S5. Rutherford E.	Baylina N. + Quirós-Pozo R.	Evaluating bottlenecks to Great Lakes Alewife <i>Alosa pseudoharengus</i> recruitment occurring in the larval stage	
12h15-12h35	S2. Quirós-Pozo R.	Baylina N. + Quirós-Pozo R.	Evaluation of different taurine levels on the weaning of the greater amberjack (<i>Seriola dumerili</i>).	
12h35-12h55	S2. Schmidt N. *	Baylina N. + Quirós-Pozo R.	How to like it less salty: Acclimatizing laboratory-reared hatchling cod (<i>Gadus morhua</i>) to salinity conditions in the Baltic Sea	
12h55-14h15	Lunch Break			
14:15 - 14:50	Plenary, Leis J.	Faria A. + Garrido S.	Dispersal-relevant behaviour of fish larvae – measuring it, putting it into models and using a dispersal model incorporating behaviour	
14h50-15h10	S2. Barraca C.	Baylina N. + Quirós-Pozo R.	Marine teleost breeding at Oceanário de Lisboa	
15h10-15h30	S2. Castanho S.	Baylina N. + Quirós-Pozo R.	Feeding different marine fish larvae species in aquaculture	
15h30-15h50	S2. Oz I. *	Baylina N. + Quirós-Pozo R.	Non-uniform metamorphosis underlies different development trajectories in hatchery-reared flathead grey mullet (<i>Mugil cephalus</i>)	
15h50-16h10	S2. Diogo P.	Baylina N. + Quirós-Pozo R.	ALLARVAE: new microalgae live feed enrichment for marine fish larvae nutrition	
16h10-16h30	Coffee Break			
16h30-18h00	Early Career Event (Room 1)			
16h30-18h00	ELHS Business meeting (Aud 2)			
18h00-18h20	Tribute to John Blaxter (Aud 2)			
18h20-21h00	Poster Session			

Wednesday, 10th May				
	SESSION/AUTHOR	CONVENERS	TITLE	
8h00-16h00	Registration desk available			
9h00-9h35	Plenary, Fuiman L.	Faria A. + Garrido S.	How Did We Get So Far From Where We Began? Looking Back on Our Team's Research on Maternal Effects	
9h35-9h55	S4. Catalan I.	Koeningshtein S. + Machado A.	Long-term interannual variability in larval dispersal and connectivity of anchoveta (<i>Engraulis ringens</i>) in the southern Humboldt system: when global events matter	
9h55-10h15	S4. Berenshtein I.	Koeningshtein S. + Machado A.	Proposed network of Marine Protected Areas substantially support larval dispersal and connectivity in the Eastern Mediterranean	
10h15-10h35	S4. Machado A.	Koeningshtein S. + Machado A.	Sardine larvae dispersion patterns and survival on the Iberian Current System	
10h35-10h55	S3. Garrido S.	Garrido S. + Moyano M. + Ré P.	The big unknowns on the feeding dynamics during the early life stages of small pelagic fishes	
10h55-11h15	Coffee Break			
11h15-11h35	S3. Kanaya A.	Garrido S. + Moyano M. + Ré P.	Comparison of the strength of hatchling size and environmental factors affecting feeding intensity of walleye pollock larvae at first feeding stage	
11h35-11h55	S3. Fiksen Ø.	Garrido S. + Moyano M. + Ré P.	Bluefin tuna spawn in an ocean desert with few predators and just enough food for first feeding larvae	
11h55-12h15	S3. Moyano M.	Garrido S. + Moyano M. + Ré P.	Shedding light into the larval diets of North Sea herring and Peruvian anchovy using metabarcoding	
12h15-12h35	S3. Murphy H.	Garrido S. + Moyano M. + Ré P.	Identifying the drivers of early life survival of Capelin (<i>Mallotus villosus</i>) in Newfoundland, Canada	
12h35-12h55	S3. Fonseca P.	Garrido S. + Moyano M. + Ré P.	Molecular barcoding reveals patterns of egg predation in small pelagic fish	
12h55-14h15	Lunch Break			
14:15 - 14:35	S5. Robert D.	Baptista V. + Teodósio A.	Life in the fast lane: revisiting the fast growth – high survival paradigm during the early life stages of fishes	
14h35-14h55	S5. dos Santos Schmidt T.	Baptista V. + Teodósio A.	Influence of temperature on density and growth of capelin (<i>Mallotus villosus</i>) larvae around the Icelandic coast	
14h55-15h15	S5. van Damme C.	Baptista V. + Teodósio A.	Improving Larvae Recruitment Survey Indices: A case study of North Sea Herring	
15h15-15h35	S5. Ferreira A.S.	Baptista V. + Teodósio A.	Ocean-climate conditions and spawner biomass affect blue whiting survival	
15h35-15h55	S1. Teletchea F.	Baptista V. + Teodósio A.	Why do fish larvae hatch when they do?	
16h00 - 18h30	Visit to Oceanario de Lisboa			
18h30-22h00	Social Dinner			



Thursday, 11th May				
	SESSION/AUTHOR	CONVENERS	TITLE	
8h00-16h00	Registration desk available			
9h00-9h35	Plenary, Robert D.	Faria A. + Garrido S.	The importance of prey selectivity for revealing relationships linking prey availability to larval feeding success and survival potential	
9h35-9h55	S5. Sirois P.	Baptista V. + Teodósio A.	Spawning habitats created to promote recruitment of a key forage fish species in a large boreal reservoir	
9h55-10h15	S5. Paris C.	Baptista V. + Teodósio A.	Larval swimming and orientation behavior of the amphidromous goby, <i>Sicydium</i> spp., of São Tomé Island	
10h15-10h35	S5. Primo A.L.	Baptista V. + Teodósio A.	Climate-driven changes of ichthyoplankton communities in an Iberian estuary – a 13 years study	
10h35-10h55	S7. Cumplido N.	Baptista V. + Teodósio A.	Ontogeny and morphology of the body axis of the Chilean Anchovy (<i>Engraulis ringens</i>)	
10h55-11h15	Coffee Break			
11h15-11h35	S1. Réalis-Doyelle E.	Baumann H. + Faria A. + Ribeiro F.	Synergistic effects of parental PCB contamination and climate change on a cold stenothermic fish	
11h35-11h55	S1. Cruz J.	Baumann H. + Faria A. + Ribeiro F.	Anthropogenic environmental stressors may change fish larvae community in a temperate river estuary	
11h55-12h15	S1. Muller C.	Baumann H. + Faria A. + Ribeiro F.	An eco-physiological investigation of fisheries-induced evolution and climate change	
12h15-12h35	S7. Miskiewicz T.	Baumann H. + Faria A. + Ribeiro F.	The identification of flathead larvae (F. Platycephalidae) and their seasonal and spatial distribution off south-eastern Australia	
12h35-12h55	S7. Stern N.	Baumann H. + Faria A. + Ribeiro F.	Revising a lost knowledge: ichthyoplankton communities in the Israeli Mediterranean coast	
12h55-14h15	Lunch Break			
14:15 - 14:35	S1. Rodrigues D.	Baumann H. + Faria A. + Ribeiro F.	Assessing the effects of nonylphenol and microplastics in <i>Sparus aurata</i> L., 1758 larvae: prey ingestion and dermal uptake pathways	
14h35-14h55	S1. Lima A.	Baumann H. + Faria A. + Ribeiro F.	Multiple responses of fish larvae to heat and hypoxia as interacting stressors: acclimation potential to extreme conditions	
14h55-15h15	S7. Thompson A.	Baumann H. + Faria A. + Ribeiro F.	Biogeography in the California Current Ecosystem from a Larval Fish Perspective	
15h15-15h35	Closing remarks			



POSTER CONTRIBUTIONS

NOTES: Please use the poster number to identify your poster slot. * Stands for candidate to the best student poster.

Poster Session	Code	First author	Title
S1. Global change effects on early life stages	PS1.1	Almeida J*	Effect of temperature on routine behavior in early life stages of gilthead seabream, <i>Sparus aurata</i>
	PS1.2	Ehlers S*	Vulnerabilities of Northwest Atlantic Living Marine Resources to Ocean Acidification
	PS1.3	Frommel A	Air exposure moderates effects of ocean acidification in early development of Pacific herring
	PS1.4	Ribeiro L	How different rearing temperature influence embryonic development and fatty acid utilisation of white seabream (<i>Diplodus sargus</i>) early life stages
	PS1.5	Rosenberg S	How extreme cold and warm oceanographic events influence larval fish assemblages in the southern region of the California Current off Mexico
	PS1.6	Tavares S*	Immune responses of juvenile <i>Diplodus sargus</i> exposed to a marine heatwave
	PS1.7	Garrido S	Effect of temperature on European sardine larval growth, condition and survival throughout ontogeny
S2. Recent trends in marine Larviculture	PS2.8	Chambers C	Identifying optimal conditions for early life-stage production of a cold-water wrasse <i>Tautoga onitis</i> : spawning, thermal regimes, culture density, and diet
	PS2.9	Fey D	Experimental and Breeding Centre at the NMFRI – new opportunities for the research on early life history of fish
	PS2.10	Gavaia P	Optimizing feeding protocols for zebrafish larvae: effects of co-feeding with live prey and microdiets on growth and skeletal deformities
S3. Larval Trophic Ecology	PS3. 11	Garrido S	Diet of Small Pelagic Fish larvae – insights from metabarcoding of larval stomach contents
S5. Larval survival and recruitment variability	PS5.12	Angélico M	Do sardine and anchovy larvae share the same spots? <i>Sardina pilchardus</i> and <i>Engraulis encrasicolus</i> larvae distributions in NW Iberian waters
	PS5.13	Ishikawa T*	Importance of growth history at first feeding stage for development and survival of larval yellow goosfish verified in laboratory experiment
	PS5.14	Peres H*	Larval retention and recruitment of small pelagic fish in a boreal fjord



	PS5.15	Szkudlarek-Pawelczyk A	Larval herring (<i>Clupea harengus</i> L.) abundance in the Pomeranian Bay (Baltic Sea) during 2007-2015 and 2019-2022
	PS5.16	Tomczak M	Effects of the vernal thermal bar on water quality, density and distribution of plankton and fish larvae in Lake Michigan, USA
S6. Application of “omics” in larval research	PS6.17	Dias D*	Using morphology and molecular techniques to describe fish larvae communities in west-African insular mangroves
	PS6.18	Azevedo O	Old meets new: 1. Morphological analyses of Guadiana ichthyoplankton as a support for DNA-based approaches
	PS6.19	Machado C	Old meets new: 2. eDNA metabarcoding vs morphological analyses for detecting ichthyoplankton in the Guadiana estuary
	PS6.20	Otorgust S*	First steps in exploring deep-sea fish larval development in the eastern Mediterranean Sea
S7. Other contributed papers	PS7.21	Andrade L	Morphological description of <i>Pogonias courbina</i> (Lacepède, 1803) larvae
	PS7.22	Costa I	Zebrabloom: novel microalgae product for zebrafish nutrition
	PS7.23	Martins-Cardoso S*	Do early life stages of fish show signs of personality?
	PS7.24	Miskiewicz T	Comparison of the juvenile morphology and assessment of ontogenetic changes in morphology of three species of seahorses <i>Hippocampus</i> spp. from south-eastern Australia
	PS7.25	Moreno A.	Drivers of sardine larvae survival in the northern Canary upwelling system
	PS7.26	Santos R	Ichthyoplankton spatial variability in the SW Atlantic during the relaxation phase of coastal upwellings



ABSTRACTS

ORAL CONTRIBUTIONS

NOTES: * stands for candidate to the best student oral communication.

S1. Global change effects on early life stages

Anthropogenic environmental stressors may change fish larvae community in a temperate river estuary

Joana **Cruz**^{1,2}, Olga M. Azevedo¹, Inês Rodrigues¹, Luciano de Oliveira Junior³, Erwan Garel³, Vânia Baptista¹, and M. Alexandra Teodósio¹

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River estuaries are important nursery areas where fish larvae can find suitable food conditions and refuge areas. However, many estuaries are subject to several anthropogenic environmental stressors, from dams' construction to plastic pollution. This study aimed at describing the ichthyoplankton community of Guadiana estuary, 16 years after the Alqueva dam closure, as well as determining the microplastics (MPs) availability, by investigating the ratio between fish larvae and MPs. Sampling occurred in spring 2018 (April to June), where 82 zooplankton tows were conducted during low/high tides along the salinity gradient of the estuary. A total of 667 fish larvae were collected, with a mean density of 35.6 larvae 100 m⁻³. The family Gobiidae presented the highest densities (43.9%) followed by Blenniidae (18.5%), with high dominance of *Pomatoschistus minutus* and *Parablennius pilicornis*. *Sardina pilchardus* and *Engraulis encrasicolus* represented only 2.2 and 2.1%, respectively. There was a relevant decrease in *E. encrasicolus* larval density, when comparing to results found before (Spring 1988) and during the Alqueva dam closure (Spring 2002), with percentages > 90%. Several studies have hypothesized that freshwater inputs alteration or invasive jellyfish blooms may explain those changes in the area, but none analyzed MPs. Microplastics were found in all samples, with an average density of 8509.2 MPs 100 m⁻³. Most MPs were smaller than 1 mm, and fibers represented 97.7%, while fragments only 2.3%. The average ratio found was 1 fish larva: 300.9 MPs, which is considerably high and probably enhances a higher risk of MPs ingestion by fish larvae.



Quantitative species-level characterization of a larval community reveals narrow thermal affinity of invasive larvae, manifested across temporal and spatial scales

*Michaela **Kolker**¹, Jonathan Belmaker^{1,7}, Moshe Kiflawi^{2,3}, Shahar Malamud¹, Tamara Gurevich^{3,4}, Yochai Meir⁵, Itai Sharon⁵, Rotem Sorek⁶, Shai Meiri^{1,7}, and Roi Holzman^{1,3,7}

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⁷ Steinhardt Museum of Natural History, Tel Aviv University, Tel Aviv, Israel.

The eastern Mediterranean Sea is on the frontier of invasion by nonindigenous Indo-Pacific species, facilitated by the opening of the Suez Canal and exacerbated by climate change. The exploration of the shifts in fish community structure that followed the invasion focused on adult stages. The difficulties in accurately identifying fish larvae down to the species-level compounds this gap in our knowledges on invasion dynamics. We addressed this gap using high-throughput Next-Generation Sequencing (NGS) to provide a quantitative, whole-community, species-level characterization of the larval assemblage from the Mediterranean coastal waters of Israel.

We found that larvae of native species appeared throughout the year and were depth-generalists, indicating a broad thermal niche. Non-native species, on the other hand, were found mainly in the upper layers under stratified conditions, indicating preference for warmer waters. This disparity extended into seasonality, where native species experienced vast community turnover in winter, whereas nonnative species larvae were almost completely absent from winter assemblages. We found that both the relative larval abundance and species richness of the non-native species increased from accounting for 8% of species and 15% of larvae in the winter, to over 35% of species and 53% of larvae at the end of the stratified period in the fall.

Our study implies that the progressively warmer waters of the Mediterranean Sea benefit disproportionately the proliferation of nonindigenous larvae. This can have far-reaching consequences for the fish community, from the establishment of novel invaders, shifting species composition, to rapid range expansion into the western basin.



Multiple responses of fish larvae to heat and hypoxia as interacting stressors: acclimation potential to extreme conditions

André Ricardo de Araújo **Lima**^{1,2}, Emily Booms³, Ana Rita Lopes^{1,4}, Sara Martins-Cardoso^{1,2}, Sara C. Novais^{1,5}, Laura Ribeiro⁶, Sara Castanho⁶, Ana Candeias-Mendes⁶, Pedro Pousão-Ferreira⁶, and Ana M. Faria^{1,2}

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⁵ ESTM, Polytechnic of Leiria, Peniche, Portugal.

⁶ Portuguese Institute for the Sea and Atmosphere - IPMA, Aquaculture Research Station, Olhão, Portugal.

Ocean's characteristics are changing at global-scale, shifting metabolism processes and the match-mismatch of sensitive early life stages within their environmental optima. Therefore, to assess the effects of warming and hypoxia on larval ecology is still crucial to predict the future of fish stocks and diversity. Here we assessed whether the individual chronic effects of warming (24°C) and hypoxia (2.5 mg L⁻¹ DO) can be amplified by the combination of these two stressors on growth, behavior (boldness, exploration and activity levels), metabolic rate, oxidative system and energy metabolism system of white seabream larval stages (*Diplodus sargus*). Our results showed that faster larval growth in length and slower gains in weight are induced by heat stress. Warming and hypoxia did not directly affect boldness and exploration, while larval activity is enhanced by heat stress and hypoxia or their combination. Larvae also decreased oxygen consumption when exposed to both stressors in combination. Moreover, variability in oxidative and energy metabolism systems significantly influenced changes in all target responses. The antioxidant system acts to reduce reactive oxygen species due to a clear balance between superoxide dismutase and catalase activities in all treatments, but did not prevent damages to DNA and to cellular membranes (lipid peroxidation) in the hypoxia treatment. Hypoxic stress also decreased the aerobic metabolism and, consequently, the cellular consumption of oxygen. We conclude that larvae can improve physiological performance to reduce direct negative effects on growth, activity and metabolism, indicating signs of acclimation to extreme conditions after chronic exposure.



How is the plastic pollution situation in the Douro Estuary? Monitorization of microplastics and larval fish assemblages in a Portuguese urbanized estuary

*Sabrina Rodrigues Magalhães^{1,2}, Diogo M. Silva^{1,2}, Rúben Pereira^{1,2}, Francisca Espincho^{1,2}, Michael Elliott³, C. Marisa R. Almeida^{1,4}, and Sandra Ramos¹

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The initial stages of fishes are vulnerable to several factors, including anthropogenic pressures. Larval fish of several marine species use estuaries as a nursery ground to improve their survival. Recent studies showed that plastic pollution is increasing in estuaries posing another pressure to these organisms. Microplastics, due to their small, pose a higher risk to fish larvae, since they can ingest them affecting their growth and survival. Due to their potential negative effects, it is important to continuously monitor microplastic contamination in aquatic environments. The Douro Estuary (NW Portugal) is a highly urbanized estuary and a previous study showed a microplastic concentration significantly higher than expected surpassing larval fish abundance. Hence, this study aimed to monitor the availability of microplastics to fish larvae in Douro Estuary. Monthly sampling surveys were performed between 2021-2022 along the axial gradient of the estuary using sub-surface planktonic trawls to collect fish larvae and microplastics. A total of 143 samples were collected; fish larvae were identified and microplastics were quantified and characterized. Microplastics were found in all the samples and were mainly composed by particles and fibers. More than 1000 fish larvae were collected and identified. Common goby, *Pomatoschistus microps*, Clupeidae n.i. and *Solea senelagensis* were the most abundant taxa. The temporal and spatial co-occurrence of microplastics and fish larvae is being analyzed. Data collected will be very relevant to indicate the bioavailability of microplastics to fish larvae and assess microplastics' ecological risks to such important organisms.



An eco-physiological investigation of fisheries-induced evolution and climate change

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The early life stages of fish are highly sensitive to environmental changes, including ocean acidification (OA). Previous studies have shown that the impacts of OA on these stages can be negative, neutral, or positive, and are often species-specific. However, there are several confounding factors, such as the life-stage examined and population history, that may influence these results. In this study, adult roman seabream *Chrysoblephus laticeps* from two populations with different histories of exploitation were used to examine the effects of heritability and OA on larval metabolism and growth up to the flexion stage. The results showed that the effects of OA on larval roman seabream were stage-specific, with preflexion larvae showing metabolic and growth depression, while flexion larvae exhibited significantly elevated metabolic rates. Offspring from the protected population, however, exhibited greater resilience to the same experimental OA conditions. These larvae did not show evidence of metabolic or growth depression, and while metabolic rates were elevated at flexion, they were considerably lower than those of offspring from the exploited population, indicating a greater degree of tolerance to an environmental stressor. The study concludes that spatial protection from exploitation may provide a means for ensuring more resilient fisheries populations in a rapidly changing ocean environment, with the genetic and phenotypic diversity necessary for adaptation to a changing environment.



Will climate change impact the Baltic cod?

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Regarding the Baltic Sea, future climate models predict an increase in water temperature and a decrease in salinity from current environmental conditions. Thus, multiple marine organisms will be forced to face and cope with these environmental changes in the near future. The cod (*Gadus morhua*), an economically and ecologically important species is one of them. During embryonic development, both temperature and salinity play a role in their viability. Cod eggs and larvae are sensitive to changes in these environmental parameters during early development and they can determine the survival of both eggs and larvae. In this study, I am to explore how well the predicted temperature and salinity conditions in the near future impact the development and performance of Baltic cod during early life stages.

In order to do this, I incubated cod eggs under current and future environmental conditions and followed larvae survival, hatching rate, and buoyancy. According to the results, temperature increase and salinity decrease will negatively impact the survival and hatching of cod larvae, moreover, buoyancy will also be affected by these parameters.

I conclude that the predicted temperature and salinity conditions will impact Baltic cod during early life development, affecting survival, hatching and buoyancy. Consequently, results suggest that under these environmental predictions, climate change will be a threat to the Baltic cod in the close future.



Climate-driven changes of ichthyoplankton communities in an Iberian estuary – a 13 years study

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Ichthyoplankton assemblages are key components of estuaries worldwide, playing a vital role as nurseries for fish larvae. Nonetheless, estuaries can be highly affected by ongoing climate change. Impacts of climate variability on ichthyoplankton assemblages will have consequences for marine pelagic food webs and fish populations biology, namely recruitment. This study aims to investigate the influence of environmental variability on the interannual abundance of ichthyoplankton assemblages of the Mondego estuary (Portugal). For this, an ichthyoplankton sampling program of 13 years (2003-2015) along six distinct sampling stations was analyzed to evaluate spatial, seasonal, and interannual changes in ichthyoplankton distribution over periods of wet, regular, and dry conditions. The ichthyoplanktonic community was dominated by *Pomatoschistus* spp. across all seasons and conditions, with higher larval abundances during summer and spring. The main changes were related to species seasonality and phenology as well as an increase in the number of marine species during extreme events. The larval fish community showed a strong relationship with the regional and local environment over the study, presenting a distinct yet highly variable structure during the 2009-2013 period. Reported changes will likely trigger major changes in species dominance and abundance, with clear ecological and socio-economic implications.



Synergistic effects of parental PCB contamination and climate change on a cold stenothermic fish

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The end of the XXth century is characterized by rapidly changing aquatic ecosystems due to the effect of human activities via the increase of multiple stresses. Among these stresses, polychlorinated biphenyls (PCBs) are bioaccumulated by aquatic fauna, and particularly fish, as a result of their lipophilic nature and their low degradation. In addition, in the present context of climate change, temperature variations may have indirect abiotic consequences on PCBs, making them more bioavailable. Arctic char is a cold stenothermic salmonid whose population in Lac du Bourget has remained at a low level for the last decade.

Thus, the aim and originality of this project is to study the intergenerational effects of parental PCB contamination under the influence of small temperature increases via a multiparametric and multiscale approach allowing the development of methods to investigate both omic and physiological mechanisms of action.

The results of our study show a population collapse due to maternal contamination coupled with an increase in temperature (12% survive maternal contamination Vs 50% survive paternal contamination). Parental transmission of PCBs affects the rhythm of life of larvae; the effects of maternal transmission being greater than the effects of paternal transmission. Indeed, synergistic effects (temperature and transmission of PCBs) have been shown on the same parameters for male and female, but additive effects of PCBs and temperature are more important, which could explain for women why the effect of maternal transmission coupled with an increase in temperature is more negative. In addition, PCBs are historical pollutants, they are POPs, so we can make the strong assumption that these results can be transposed to other POPs.



Evaluating bottlenecks to Great Lakes Alewife *Alosa pseudoharengus* recruitment occurring in the larval stage

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Alewife is a non-native fish that supports economically valuable Great Lakes salmonine fisheries, and has recently declined owing to variable piscivore predation on adults and poor recruitment. Factors known to influence Alewife recruitment include salmonine consumption of adults, warm water temperatures, and lower wind speeds during spring and summer when larvae are present. We hypothesized that Alewife recruitment variability also is related to aquatic invasive species effects. Specifically, since 2004, filtration by the invasive quagga mussel *Dreissena bugensis* has increased water clarity, shunted nutrients from pelagic to benthic pathways, and lowered productivity, biomass and altered composition of zooplankton prey. From 2010-2021, we conducted fine-scale spatial surveys of water quality, plankton and larval fish at nearshore, mid-depth and offshore sites during June and July in Lake Michigan, and compared Alewife densities, diets, and otolith-derived growth and survival rates to pre-quagga invasion estimates. We found Alewife larval mortality rates were lower than before *Dreissena* mussels. Although mean densities of Alewife larvae have not declined since mussel invasion, variance has increased. In most years, young Alewife larvae consumed mussel veligers, grew poorly, and experienced several upwelling events that displaced them from warmer nearshore sites to colder offshore sites. However, in 2010, water temperatures were warm and led to early Alewife spawn and hatch dates, larvae experienced no upwellings, consumed fewer mussel veligers because of their rarity in the plankton, and subsequent recruitment was high. Our results suggest upwellings and poor prey composition during the larval stage are additional bottlenecks to successful Alewife recruitments.



Legacy effects of variation in larval size at hatch: A case study of California Grunion

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Previous studies on larval fishes have helped scientists conclude that offspring size is often correlated with quality. However, mechanistic assessments of the relationship between offspring size and performance are relatively rare, and the degree to which environmental conditions may affect these relationships is unclear. In a study of California Grunion, we investigated the relationship between larval size at hatch (SAH) and several performance metrics including rates of feeding, metabolism, growth, and mortality. The performance of larvae was measured in an experiment where ambient and high pCO₂ treatments were crossed with low and high ration treatments. Generally speaking, SAH varied substantially across both individuals and families and was positively correlated with post-hatch performance. SAH was positively correlated with subsequent sizes at 6 dph and 12 dph as well as specific growth rates between 6 dph and 12 dph. SAH is also negatively correlated with mortality rates, where families with larger larvae had lower mortality rates across treatments. Finally, larger SAH grants some resistance to the effects of ocean acidification, particularly when investigating feeding rates. Larvae in families that were smaller at hatch had lower feeding rates under OA conditions than under ambient conditions, while families where larvae were larger at hatch had feeding rates under OA conditions that were comparable to those under ambient conditions. Understanding how initial size and different physiological traits co-vary under climate change conditions is important as we strive to understand the mechanisms that moderate larval fitness under climate change.



Why do fish larvae hatch when they do?

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Hatching, which corresponds to the moment an individual leaves its egg envelopes (chorion), is the result of a softening of the chorion caused by two proteases secreted by the hatching glands, as enhanced by the embryo's increasing activity. Also, the time of hatching, which strongly varies between species, is environmentally controlled, with temperature and oxygen concentration exerting the major effect. Yet, to our knowledge, nobody ever explained why fish larvae hatch when they do. Based on the Gill-Oxygen Limitation Theory (GOLT), originally developed for later stages of fish, we tested whether larvae hatch when a specific ratio between egg surface area (ESA) and larval body surface area (LBSA) is reached, based on the hypothesis that hatching occurs when supply of oxygen within the egg becomes insufficient, so that larvae must hatch or die of hypoxia. For this proof-of-concept study, we assembled information for five marine species. We found a strong relationship: $LBSA = 0.80 \text{ EAS}$ ($r^2 = 0.96$). This result implies that hatching occurs when larvae reach a certain size within its egg shell. In other words, it is the lack of oxygen within the egg shell that triggers both the secretion of proteases and the increasing activity of the larvae. Because the above LBSA/EAS ratio is based on only five species, our hypothesis needs to be tested with a larger dataset. These results are pertinent to global warming and deoxygenation, which may cause premature hatching.



Co-exposure of tire wear particle coupled with increased temperature during the early life stages of a cold stenothermic lake fish, the char

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The global anthropization of lakes shorelines stresses the ecosystems in diverse manners. For instance, Lake Geneva, one of the greatest lakes of western Europe located on the north side of the Alps, has witnessed rising plastic pollution in its waters, estimated between 33000 and 220000 particles/km², of which about 60% has been identified as tire wear particles (TWP). The TWP contain a degradation product from an anti-ozone molecule, N-(1,3-dimethylbutyl)-N'-phenyl-p-phenylenediamine-quinone (6PPD-Q), recently reported as very highly toxic on coho salmon (*Oncorhynchus kisutch*). This study focuses on the sublethal effects of 6PPD-Q coupled with an increase of temperature on early life stages in arctic charr (*Salvelinus alpinus*), a cold stenothermic Salmonidae of which the subalpine lakes constitute the southern bound of its natural range.

The larvae have either been contaminated once, at the embryonic neuronal development stage or several times when they reached a critical ecological stage. Several traits are currently being measured to assess an effect upon behavior alongside morphological traits. The behavioral traits will be characterized as average and maximum speed, aggressiveness, swimming activity, covered distance and response to the introduction of a potential predator. The first results seem to indicate an increase in malformations of larvae linked with the temperature and the presence of the toxic compound.



Variability in fish phenology over 20 years of rapid warming along the Northeast US Continental Shelf

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Change in phenology is one of the hallmarks of global climate change. In fish, marine warming is expected to cause the advancement of a spring peak in larval occurrence and/or the delay of a fall peak. However, empirical evidence has not consistently upheld this broad prediction, implying that more research, and perhaps more nuanced hypotheses, are needed. Our study investigates warming impacts on fish phenology by examining patterns in larval occurrence on the Northeast US Continental Shelf, one of the most rapidly warming regions of the global ocean. We use data from NOAA's expansive Ecosystem Monitoring program, which has been sampling shelf-wide, cataloging dozens of species of larvae, and recording in situ temperature and salinity on a bimonthly basis since 1999 – prior to the most dramatic contemporary warming. We calculate the central tendency of seasonal larval occurrence for 38 species and test for common drivers influencing changes in larval seasonality. Our analyses fail to provide evidence for consistent trends in larval timing over the last two decades, nor do we see clear patterns based on adult habitat. Rather, we find that variability in larval timing shifts across species and space, with the shelf break region showing the highest degree of interannual variation. Our results suggest that uniform or linear phenological responses to warming are unlikely. Instead, understanding past and present variability in larval occurrence, and how it differs based on habitat, can yield fresh insights into how fish populations have and will continue to respond to climate change.



S2. Larviculture

Marine teleost breeding at Oceanário de Lisboa

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Marine fish breeding is a challenge and the number of species successfully reared is currently limited, however this number has been increasing in the last years. The successful breeding of marine species could contribute to mitigate the consequences of overfishing or habitat destruction that are threatening a lot of wild populations. Public Aquariums are playing an important role on the study of a diversity of species as they offer great opportunities to watch behaviors and obtain samples and information for species that are difficult to access in the wild. Modern public aquariums due to their dimension and multispecies displays create very good conditions that promote marine teleost breeding behaviors. Oceanário de Lisboa has been focusing in the last years on developing the techniques to collect the eggs and raise the larvae of several different species. The aim of this presentation is to describe the work that has been done by the Oceanário Breeding Research Team including the main challenges, bottlenecks and results.



Feeding different marine fish larvae species in aquaculture

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Sustainable production of marine fish in aquaculture is highly dependent on the production of high-quality larvae. Understanding the behavior, physiology, and metabolism at different larval developmental stages, in rearing conditions, contributes to optimize feeding protocols, enhances growth and survival, but can also provide insights into ecological studies. When developing feeding protocols, feed must be appropriate to the mouth gap, adequate buoyancy (inert diet), adapted to swimming ability, nutritionally adequate, and adapted to digestive capacity. Live feeds, such as rotifers (*Brachionus* spp.) and Artemia (*Artemia* sp.), are not the natural prey of marine fish larvae, but are commonly used in aquaculture do to their ease to produce at high densities, short life cycle (rotifers), resistant eggs that can be hatched on demand (Artemia), appropriate size for the mouth gap, slow swimming adequate for larvae locomotion abilities, and the possibility to enrich with nutrients according to larval nutritional requirements. Inert microdiets are easy to use and maintain a constant nutritional profile. However, they still cannot completely replace live feeds. Typically, feeding sequence for marine fish larvae progresses from rotifers to Artemia, and finally inert diets, but the protocol varies depending on the cultivated species. For instance, seabass larvae are only fed after they inflate their swim bladder and not since mouth opening, unlike other species. Seabass and sole, can start feeding on both rotifers and Artemia while seabream start feeding with Artemia two weeks later due to their mouth gap size. Concluding, feeding protocols must be fitted to the cultivated species. This work had the support of the project DIVERSIAQUA II (Mar2020-P02M01-0656P).



ALLARVAE: new microalgae live feed enrichment for marine fish larvae nutrition

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Development of new products based on the combination of microalgae and probiotics is a valuable formulation strategy to support fish nutrition in early life stages. Different microalgae species can be used in formulations to meet specific nutritional requirements, and inclusion of probiotics may support the improvement of larvae resilience to stressful factors. ALLarvae project focused on the development of microalgae products in powder with probiotics for hatcheries: ALLgreen (green water technique), ALLive (live feed nutrition) and ALLarvae (live feed enrichment). Two *Nannochloropsis* sp. liquid concentrates were also developed with low bacterial content for rotifers nutrition production in closed systems (Phytobloom® ELITE formula) and in Recirculation Aquaculture Systems (Phytobloom® RAS formula). In hatcheries there are necessary stressful factors that promote mortality such as larvae transport, which is particularly disadvantageous in marine species with low survival in early life stages. ALLarvae pilot formulation was manufactured and tested according to characteristics such as decantation, microalgae agglomerates abundance and dimensions and biochemical profile. Seabream (*Sparus aurata*) were cultured until weaning with commercial products or pilot ALLarvae formulation (*Nannochloropsis* sp., *Tisochrysis* sp., *Aurantiochytrium* sp., *Tetraselmis* sp., probiotics and vitamins) as enrichment in triplicate. Larvae growth and water quality was evaluated. Control group revealed 15.5% of survival while the ALLarvae treatment revealed 18.7%. A stress challenge of transport (220Km) was performed in the end of the experiment where control showed lower survival (75.4%) when compared to ALLarvae treatment (85%). ALLarvae formulation showed to be nutritionally adequate for seabream larvae promoting higher larvae resistance to stressful events. Acknowledgements: Supported by Project ALLarvae - 069971 supported by CRESC Algarve, Portugal 2020 and European Union.



Evaluation of different taurine levels on the weaning of the greater amberjack (*Seriola dumerili*)

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Taurine is an essential nutrient for larval development and growth, mainly in carnivorous marine fish such as the greater amberjack (*Seriola dumerili*). However, there are no available data on dietary taurine supplementation during the weaning period of this species. For those reasons, four granulated microdiets containing 0.24 to 4.24 % of taurine levels were evaluated in 30 days post-hatching (dph) greater amberjack larvae until 44 dph. Growth parameters, survival, histology, and skeletal anomalies were determined. The larvae fed Tau 1.24 had the greatest total length, weight gain and daily weight gain. Additionally, larvae fed Tau 0.24, Tau 1.24 and Tau 2.24 presented a healthy liver appearance, while the valorization of the intestine along the different regions was similar between treatments. However, total skeletal severe anomalies were high for the groups fed taurine at 0.24 and 4.24%. Therefore, the present results suggest the importance of adequate taurine supplementation in the weaning of the greater amberjack, as it impacts the growth and the incidence of skeletal anomalies in the fish.

Non-uniform metamorphosis underlies different development trajectories in hatchery-reared flathead grey mullet (*Mugil cephalus*)

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The flathead grey mullet (*Mugil cephalus*) is an important food fish and is at the focus of an intense domestication effort. To date, mullet aquaculture depends upon capture of wild fry in coastal waters and estuaries. To allow domestication of this species, much effort was invested to close mullet's life cycle in captivity. Today, the main bottleneck faced by mullet hatcheries is the non-uniform development of the larvae within the spawning batch.

We therefore aimed to identify the underlying causes for this phenomenon. As a first step, we generated a detailed staging system of mullet larval development in captivity, based upon morphological features. Using this developmental atlas to study development dynamics, we found that mullet larvae exhibit a biphasic growth profile with a 17-fold increase in growth rates at the onset of metamorphosis. Moreover, we found that within rearing batches, size variation almost doubles at metamorphosis, suggesting that the onset of metamorphosis is a critical step that increases variation by dictating different growth trajectories to early and late-metamorphosing larvae. We describe how qualitative morphometric traits develop as a function of quantitative parameters of age, length, eye diameter and weight.

This work is an important milestone in the domestication efforts of the grey mullet and will be used as a foundation to understand the genetic and hormonal landscape of larval development.



How to like it less salty: Acclimatizing laboratory-reared hatchling cod (*Gadus morhua*) to salinity conditions in the Baltic Sea

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Decades of overfishing and unsustainable management, paired with habitat degradation and eutrophication, depleted the cod (*Gadus morhua*) stocks in the Baltic Sea. Accompanying severe oxygen deficiency and decreased salinity in their spawning grounds restricted successful spawning today to the Bornholm basin, resulting in decreased recruitment. In order for the species to recover, several different measures are required, among others restocking. We therefore investigated the possibility of producing laboratory-reared cod larvae acclimatized to the changed environment in the Baltic Sea. For this, cod were reared from newly fertilized eggs to non-feeding yolk-sac larvae, testing the effect of different salinity reduction treatments during early development on mortality, hatching success, and neutral buoyancy. The results show that a sudden ambient salinity decrease after hatching has no strong effect on survival or hatching (around 60 % and 95 %, respectively), while it decreased neutral buoyancy of larvae from 18 to minimum 12.5 psu. Lowest buoyancy was reached in treatments with a salinity change shortly after hatching. Gradual salinity decrease starting early in the egg stage yielded to significantly increased mortality and reduced hatching success, but also lowest buoyancy of 12 psu. We showed that a decrease of ambient salinity enables the production of yolk-sac cod larvae with reduced buoyancy, which are potentially better acclimatized to survive in current environmental conditions in the Baltic Sea.



S3. Larval Trophic Ecology

Ecological implications of a cryptic species complex on the diet of early life stages of rainbow smelt in the maximum turbidity zone of the St. Lawrence Estuary

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Rainbow smelt (*Osmerus mordax*) uses the maximum turbidity zone (MTZ) of the St. Lawrence as a nursery area and its prey field is strongly dominated by the calanoid copepod complex *Eurytemora affinis* in this highly productive area. This cryptic species complex is considered an important link between primary production and higher trophic levels and represents a crucial prey for fish larvae. However, no data is available on the trophic role of each of the cryptic species, so we aimed to evaluate the contribution of *Eurytemora affinis* and *E. carolleeae* to the diet of early stages of rainbow smelt throughout the heterogeneous habitat mosaic of the MTZ. Four different surveys were carried out during the summer of 2021 (June-August), sampling a portion of 100 km, from Quebec City to Baie Ste-Anne. The distribution of smelt and its copepod prey was heterogeneous with highest mean abundances found in oligohaline habitats (0.5-5.0 PSU; 20.41 ± 10.82 larvae 100 m⁻³). Ontogenetic diet shifts were observed, *Eurytemora* spp. being the predominant prey in June and July (75.80 – 92.74%), while the mysid *Neomysis americana* increased in importance in August (80.02%, oligohaline habitats). Using a newly developed qPCR assay, we revealed for the first time that *E. affinis* was the predominant prey with 71.9% of stomachs presenting only this species, thus *E. carolleeae* played a minor role in energy transfer. Similarly, *E. affinis* in the environment dominated meaning the rainbow smelt larvae are exploiting the most abundant resource available.



Molecular barcoding reveals patterns of egg predation in small pelagic fish

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Cannibalism and intraguild predation occur in a vast number of small pelagic fish (SPF) species. Egg and larval predation have important consequences on mortality, and its accurate assessment is important to estimate the impact on recruitment strength and population dynamics, of both predators and prey. Such assessments are hampered by the inability to visually identify the species of many fish eggs and larvae in the stomachs. Here we perform a molecular identification of fish eggs from stomach contents of European sardine (*Sardina pilchardus*) and Atlantic chub mackerel (*Scomber colias*), the dominant species of the pelagic food web off the Canary Current Upwelling ecosystem and major predators of fish eggs. Molecular identification was based on DNA barcoding, combining Sanger sequencing of individual eggs with high throughput sequencing of multiple eggs on a per stomach basis.



The big unknowns on the Trophic Ecology of small pelagic fish early life stages

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Early life stages of fish constitute a bottleneck for most fish populations, particularly for small pelagic fish for which the interannual variability of recruitment strength is very high and recruits frequently constitute the bulk of the population biomass. Finding the right prey (in terms of size and quality) during these early stages is critical for larval survival and later recruitment success. In this work we synthesize the available literature on the diet and feeding dynamics of the early life stages of small pelagic fish, including sardines, anchovies, herrings and sprats. Gut content analysis of field-caught larvae remains the most common technique to study larval diets, but the contribution from other techniques have changed in time, with a higher contribution of biomarkers (e.g. stable isotopes and fatty acid composition), molecular tools (e.g. metabarcoding) and multitrophic approaches in the past decade. While significant knowledge has been gained in the past decade, such as valuable insights on larval feeding rates and behavior through laboratory experiments in species that have been brought into culture (e.g. Atlantic herring, Pacific and Atlantic sardine), some old challenges remain. For example, larvae of small pelagics have very high vacuity rates, complicating analysis of diets in the wild. Finally, we provide recommendations for future analysis of diets and feeding dynamics in larvae from small pelagics. Such studies are essential to better understand larval growth and survival in the sea, and thus to better understand and predict population dynamics in small pelagic fish dynamics.



Diet composition and prey selectivity of larval Winter Flounder in the southern Gulf of St. Lawrence

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Newly-hatched fish larvae must turn to live prey to survive upon yolk exhaustion. The start of exogenous feeding constitutes a critical period, with prey availability driving feeding success given their limited swimming and hunting capacities. Variability in feeding success generates variability in mortality rate, which translates into recruitment variability. This project focuses on Winter Flounder (*Pseudopleuronectes americanus*), a commercially important Atlantic coastal flatfish. Recent assessments revealed that the stock in the southern Gulf of St. Lawrence is facing very high levels of mortality and low recruitment, but the role of larval dynamics in this context remains largely unknown. To provide a better understanding of potential recruitment drivers in this declining stock, we used historical samples from the Magdalen Shallows collected in 1998 to perform a first detailed assessment of diet composition and trophodynamics. Zooplankton assemblage was resolved at a fine taxonomical level, and prey consumed by larvae were identified to the lowest taxonomic level possible, including developmental stage. Copepods were grouped into two nauplius (stages N1-N3 and N4-N6) and copepodite (CI-CIII and CIV-CVI) categories. Prey selectivity was then quantified using Chesson's α selectivity index. This

assessment revealed that larval flounder can feed on multiple zooplankton taxa, and that prey selectivity changes according to relative prey abundance. However, stages N1-N3 of *Temora* spp. and *Pseudocalanus* spp. were systematically selected by larvae, pointing to a potentially high importance of the phenology and production of mid-sized calanoid copepods for larval feeding success and recruitment.



Comparison of the strength of hatchling size and environmental factors affecting feeding intensity of walleye pollock larvae at first feeding stage

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Feeding success in the larval stage is an essential component of survival and many environmental and biological factors are known to affect directly. Walleye pollock, *Gadus chalcogrammus*, also has been studied to elucidate the fluctuations in its population, and recent studies attempt to detect the characteristic of viable individuals and relationships to growth hypothesis and maternal effect by tracing growth and hatch size based on otoliths. But the effect of hatch size and the advantages on feeding and growth remains unclear. We focused on the first feeding stage larvae (5-7 mm) collected in Funka Bay, Hokkaido, and compared the magnitude of each environmental and biological effect on feeding intensity and growth rate by using piecewise SEM analysis. As the variants in this analysis, we used 15-m-depth water temperature and density of copepods nauplii (PD: prey density), global solar radiation, investigation time, notochord length (NL) and hatch check diameter of otolith (HCD) which is positively related to hatch size. The feeding intensity was significantly affected by NL, temperature and PD, but not by HCD. The effect magnitude of temperature and PD had almost half that of NL (Standardized coefficient: 0.18, 0.15 and 0.37 respectively). In addition, growth rate was not directly affected from temperature, feeding intensity and HCD. The present study revealed that advantage of bigger NL may be revoked under lower temperature and PD. Since the difficulty of the collection of individuals deceased by starvation, the true differences in hatch size might be greater than presently estimated.



Shedding light into the larval diets of North Sea herring and Peruvian anchovy using metabarcoding

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Assessing diets and prey preference of young marine fish larvae is very challenging. First, identifying organisms to the species level is difficult using morphology, especially if they are partially digested and degraded. Second, soft-bodied organisms (such as protists or jellyfish) are rapidly digested and are not well preserved in formaldehyde or ethanol. Metabarcoding is emerging as an alternative to the traditional visual identification of larval gut contents, providing a higher taxonomical resolution even on soft-bodied organisms. In this study we analyzed the larval gut contents of Atlantic herring (*Clupea harengus*) in the North Sea (September 2019) and Peruvian anchovy (*Engraulis ringens*) in the Humboldt Current System (December 2018) using a two-step metabarcoding approach (targeting protist and metazoan organisms). Larvae of both species had a diverse diet, supporting a more generalist feeding strategy. Appendicularians, dinoflagellates, hydrozoans, copepods and gastropods largely contributed to the diet of North Sea herring, while hydrozoans, ciliates, diatoms and dinoflagellates mostly contributed to the diet of Peruvian anchovy. The gut content results were then compared to the in situ planktonic community (assessed via metabarcoding and microscopy). Our results add to the growing body of knowledge that emphasize the role of gelatinous zooplankton in the diet of different marine fishes.



Identifying the drivers of early life survival of Capelin (*Mallotus villosus*) in Newfoundland, Canada

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Capelin (*Mallotus villosus*), a forage fish with a sub-arctic circumpolar distribution, collapsed in the Northwest Atlantic (NAFO Divisions 2J3KL; Newfoundland and Labrador shelves) in the early 1990s and has not recovered. Earlier research related capelin survival in the first weeks and months of life to year-class strength. The drivers of larval survival have not been identified but interannual differences in annual larval daily growth rates was found among three capelin year-classes which may be related to prey availability. This study tests the hypothesis that well-fed larvae grow faster and have improved survival. We related inshore environmental variables, zooplankton prey availability, and detailed larval diet analysis to otolith daily growth measurements for the years 2020-2022. Capelin larvae were sampled in August and September in Trinity Bay, Newfoundland. There was inter-annual variability in larval densities, with higher larval densities in 2021 (44.9 ± 18.1 ind. m^{-2}) compared to 2020 (13.9 ± 2.3 ind. m^{-2}) and 2022 (28.4 ± 1.4 ind. m^{-2}). A total of 486 stomachs were analyzed. The proportion of empty stomachs was high in 2020 (0.42) and 2021 (0.48), and lower in 2022 (0.13). Digested matter was the main prey item in all years (proportion range 0.41-0.53). Of the prey that could be identified, copepod fragments, *Oithona* spp. and *Pseudocalanus* spp. copepodites, and Calanoid nauplii were the main prey items in all three years. Otolith microstructure and zooplankton analyses are ongoing. The findings from this research will provide a mechanistic understanding of what is driving capelin productivity in the Newfoundland ecosystem.



Relationship between diet and morphology during larval development in different fish species of the Gulf of St. Lawrence

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Fluctuations in the abundance of fish populations are largely regulated by survival during the larval stage. Larval survival is highly dependent on feeding success, which in turn depends on the spatio-temporal overlap between the emergence of larval fish and the production of their preferred planktonic prey. This tight coupling between larval feeding and survival underscores the need to identify potential factors that determine the feeding preferences during a critical stage of the fish life cycle. The purpose of the present study was to study the potential influence of morphological characteristics on larval feeding selectivity. We compared the diets of 11 species of larval fish of different morphology (4 orders and 8 families) captured at the same time together with potential plankton prey. To compare the feeding niches, stomach contents of a total of 247 larvae were analyzed and ingested prey were identified to the lowest taxonomic level possible. Prey selectivity was determined by comparing the relative abundance of prey found in the stomachs to that in the environment. Despite the great morphological diversity among larval species, the strong majority of species selected the naupliar stages of the calanoid copepod *Pseudocalanus* sp.. Hence, the behavior of the larvae and their potential prey is likely more important in influencing diet composition than morphological characteristics of the predators. Our results also suggest that variability in the abundance and phenology of this key prey (*Pseudocalanus* sp.) has the potential to modulate the strength of recruitment of several fish species in Gulf of St. Lawrence.



Bluefin tuna spawn in an ocean desert with few predators and just enough food for first feeding larvae

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Atlantic bluefin tuna are large apex predators roaming the Atlantic Ocean from east to west in search for prey and could potentially spawn in any location in the Northern Atlantic. However, a key spawning ground is an ocean desert in the Mediterranean Sea. We present a theoretical argument for why this place and time for spawning has evolved, by use of an individual-based model of feeding, growth, and mortality of early life stages. The model is carefully developed alongside laboratory measurements and observations, and we use long timeseries of field data in the Balearic Sea to answer the question: Why do Atlantic bluefin tuna select just these areas for their offspring? First, the water here is warm enough for eggs to hatch and larvae to grow, but not so warm that they face a metabolic meltdown where energy needs exceed their feeding rates. Second, there is just enough food available in the warm surface layer to sustain the high growth of the first feeding larvae, allowing for short planktivore stage durations. Third, there are few predators around – and tuna also spawn at a certain time in the lunar cycle to reduce the predation risk of larvae even further. For bluefin tuna, higher temperatures can be beneficial if prey abundance is high, but critical if not. Spawning phenology and spawning location are fine-tuned to several drives including local prey or predator seasonal cycles in abundance and temperature.



S4. Integrating and modelling early life-history processes and connectivity

Proposed network of Marine Protected Areas substantially support larval dispersal and connectivity in the Eastern Mediterranean

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The marine environment of the Israeli Mediterranean is under growing threat due to natural and anthropogenic stressors. Networks of Marine Protected Areas (MPAs) are effective tools in protecting marine environments and conserving their biodiversity. Currently, only 4% of the Israeli territorial waters are declared as MPAs, however six new MPAs, which will encompass more than 20% of the Israeli territorial waters, are planned. A central component in the effectiveness of MPAs is the degree to which the protected populations are connected. The purpose of our study is performing a comprehensive connectivity analysis for the proposed network of MPAs. We find that the proposed network substantially supports local and regional larval connectivity patterns for five target species in terms of the number of recruits, betweenness centrality, as well as the number of regional and local MPAs connections. Overall, the results provide strong support for the effectiveness of the proposed MPAs in supporting local and regional larval connectivity. The framework developed as part of this study will be useful for examining other pressing ecological questions concerning natural and anthropogenic stressors such as marine pollution and invasive species.



Long-term interannual variability in larval dispersal and connectivity of anchoveta (*Engraulis ringens*) in the southern Humboldt system: when global events matter

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The dispersal of planktonic eggs and larvae is a crucial process that determines connectivity and survival probability between spawning and early juvenile suitable habitat in pelagic fish. This process ultimately affects recruitment success. In this study, we developed a biophysical model of the early life history of anchoveta by coupling an individual-based offline model with an interannual regional oceanic modeling system (ROMS). We used this model to investigate the main factors driving interannual changes in the dispersal of early stages of anchoveta with emphasis on the variability forced by El Niño-Southern Oscillation (ENSO) dynamics during the period 1994-2020. For each spawning season, individuals were released from different spawning areas along the coast and tracked for a period until they reached 20 mm in length (planktonic phase). Individuals that remained in coastal areas were considered recruited. We showed the importance of the southern spawning zone (38°-40°S) where lower wind stress, the larger continental shelf, and freshwater discharge from rivers prevent offshore advection. We also found that larval trajectories varied drastically from year-to-year following ENSO-related wind-driven transport changes. During El Niño, the probability of larval success increased by more than 30% (31%-72%), while during La Niña it decreased by less than 10% (3%-7%). We discuss the modulation of ENSO on coastal-retention and the implications for anchoveta recruitment, as well as the potential effects on larval growth and pelagic duration. Finally, we demonstrate that our biophysical simulations are coherent with existing recruitment proxies and, therefore, open new possibilities for fisheries management.



Using a numerical model to propose a new laboratory oil exposure design

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Early life stages of fish are known to be vulnerable to oil exposure, which causes both acute mortality and chronic deformations. For this reason, risk assessments of potential oil spills should include fish early life stages. This is traditionally done by establishing effect thresholds in controlled laboratory experiments with constant exposure, and then estimate the potential impact area of the spill based on the concentration thresholds. However, individual oil exposure histories could be highly dynamic, and the concentrations experienced by individuals far from constant. To improve risk assessments, it is therefore desirable to quantify the exposure experienced by individuals during a given oil spill scenario. Having this, laboratory experiments should be performed using specific exposure profiles to improve the relevance of the experiment. Finally, the experimental data could then again improve effect models for the given scenario. Here, we perform the first part of this process for a large oil spill during the spawning season of NEA cod in the Lofoten region of Northern Norway. Using a coupled numerical model with modules for ocean, oil spill and fate and fish early life stages we calculate individual exposure life histories. Based on these we propose an exposure profile that resembles the exposure of individuals during a large oil spill with long duration and high rate. The main deviating feature of this profile compared to the traditional design of constant exposure is a transient peak with high concentration occurring just when the individual hits the impact area of the oil spill.



Projecting the unpredictable: Integration of early life stage processes to quantify recruitment and forecast fish stock dynamics

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Recruitment of marine fish stocks is known to be strongly affected by environmental and ecological drivers. However, traditional stock-recruitment models have failed to improve understanding of variability in fish stock productivity and the conditions for decline and recovery, and anticipate responses to climate change, marine heat waves and other environmental phenomena.

Time-dynamic early life stage (ELS) models can improve the integration of biological rates into ecological models, bridging from empirical results to fish population and ecosystem dynamics. I will present examples of ELS models for California Current sardine and anchovy, Portuguese sardine, and Barents Sea cod. These models integrate the effects of temperature, food availability, offshore transport, pH and/or oxygen on fish early life stage processes, such as egg fertilization, egg and larval survival, growth and development. Using input from detailed regional ocean and plankton models and data, dynamic ELS models can be used to reproduce stock recruitment estimates, fit population models to survey and assessment data, and project future impacts on the stock under multiple changing environmental drivers (quantifying confidence limits based on various sources of uncertainty).

Advancing these models further will be valuable to improve interdisciplinary collaboration in fish biology, better understand climate change impacts, and develop environment-responsive fisheries management strategies, especially for novel conditions in the future.



A Fish Tale As Old As Time: Larval Abundance Is Related To Spawning Stock Size - Or Is It?

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The long-established relationship between fish larval abundance and spawning stock size provides fishery managers with an economical and feasible means of tracking the status and trends of fish stocks and can help inform stock-recruit dynamics. However, the predictive ability of larval abundance data to forecast fishery recruitment has been shown to be less suitable (or not suitable) relative to juvenile abundance, as larvae typically exhibit higher mortality due to environmental variability. Nevertheless, many important fishery species lack juvenile abundance indices and an established relationship between larval abundance and spawning stock size, or larval trends appear to lead fishery recruitment. Here, we take advantage of the uniquely long and spatially rich larval time-series data from the California Cooperative Fisheries Investigations (CalCOFI) to explore relationships between larval abundance, spawning stock size, and future fishery recruitment for select recreational fish species. We will construct standardized indices of larval abundance, total catch-per-unit-of-effort (CPUE) in the fishery (as a proxy for spawning biomass), and CPUE of fishery recruits to test causality among each using a method that implicitly accounts for lags and changing relationships (via environmental or other drivers) through time (i.e., convergent cross mapping). In doing so, we intend to demonstrate variability in these relationships across species and the added value of CalCOFI larval data as having the potential to forecast good and bad fishery recruitment years. In the face of a changing ocean, the ability to predict fishery recruitment will be ever more important for guiding sustainable fishery management, especially in data-limited contexts.



Online Model Library of Larval Fish: A Tool to Better Understand Larval Survival in Marine Ecosystems

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The stability and resilience of fish populations depends strongly on the annual recruitment of young-of-the-year. Understanding this phenomenon is crucial for the sustainable management of fish stocks. To achieve this understanding, previous studies have used individual-based models (IBMs), which simulate the behavior and interactions of individual fish during early life stages and their environment. However, information about these models is scattered across a multitude of publications and is not easily accessible to the wider scientific community. To address this issue, we compiled an online library of fish early life stages (ELSs), which brings together IBMs from various fish species, including Atlantic cod (*Gadus morhua*), Atlantic herring (*Clupea harengus*) and European sprat (*Sprattus sprattus*), among others. Our library provides comprehensive access to these models, including their summaries, codes, and quality control measures to ensure their accuracy. Our Online Library of Fish ELSs not only features models but also includes current various data for model calibration and validation. One of the goals is to develop a generic approach for larval fish IBMs suitable for various species. To achieve this, we review and compare the foraging behavior of different species to identify similarities and differences. Overall, our Online Library of Fish ELSs aims to provide a one-stop resource for researchers interested in larval fish IBMs as a powerful tool to investigate drivers of fish recruitment. By consolidating these models and data in one place, we hope to facilitate future research in this critical field and contribute to the sustainable management of fish populations.



Sardine larvae dispersion patterns and survival on the Iberian Current System

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Sardine has been the dominant small pelagic fish species off western Iberia for as long as data is recorded. Management of sardine is confounded by the high interannual variability of recruitment strength and is thought that environmental conditions experienced by the early life stages are responsible for such variability. The knowledge of the key environmental processes impacting sardine recruitment strength would allow to improve management advice. A set of different models are used to simulate the dispersion and survival of sardine early life stages in the Iberian Current System. A high-resolution simulation (IBv2.0) with the hydrodynamic model ROMS provides the oceanographic fields that are used as background for the lagrangian simulations performed with the model Parcels coupled to an Individual Based Model (IBM) of sardine eggs and larvae. The IBM is developed based on the relation between vital rates and oceanographic variables, available in the literature and obtained through laboratory-controlled experimentation, it simulates the different early life stages of sardine (egg, yolk-sac larvae, first feeding larvae) considering the effects of temperature and food availability on growth, survival, and development throughout ontogeny. The lagrangian/IBM models are used to study the dispersal of sardine eggs and larvae during the sardine spawning season off Iberian waters and the dispersal patterns and survival rates are obtained for different years with contrasting recruitment strength.



S5. Larval survival and recruitment variability

What do we know about the little fish of São Tomé and Príncipe?

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The rivers of São Tomé Island are colonized by the little fish that includes at least three species: *Awaous lateristriga* (Duméril, 1861), *Sicydium brevifile* Ogilvie-Grant, 1884, *Sicydium bustamantei* Greeff, 1884. These endemic species are amphidromous fish that spawn in those areas. After hatching, larvae drift to the ocean with the river flow. In the marine realm, the planktonic larvae develop and migrate to freshwater as post-larvae. The migrations of post-larvae support important local fisheries at the mouth of the rivers in tropical volcanic islands like São Tomé and Príncipe. However, the biology and ecology of these species in the West African islands are understudied, despite their importance for local fisheries. Thus, this study aimed to revise the knowledge regarding little fish in São Tomé and Príncipe. According to the local communities, the little fish are already in the adult phase, but analyzing the morphological characters, we know that they are still post-larvae/juveniles. When the little fish post-larvae reach the mouth of the rivers, the communities are already expecting them, since they start to appear inside the gut of big pelagic fishes previously caught by fishermen in offshore areas. The communities start catching them with “tchangas”, mosquito nets, baskets, cloths, etc. Concerning their feeding ecology, previous work showed that they are omnivorous and secondary consumers, with zooplankton and macroalgae detritus being their primary sources of organic matter assimilated. These data are only a part of the information that will be presented; more exciting details will be shown for the first time. Wait for the pictures and videos!



Ocean-climate conditions and spawner biomass affect blue whiting survival

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Blue whiting (BW, *Micromesistius poutassou*) recruitment has shown marked fluctuations since the early 1980s, with some year classes being nearly 10-fold larger than others. Hitherto, no models have accurately explained these past recruitment variations, potentially due to the lack of studies addressing BW stock dynamics in relation to large-scale oceanographic processes. Here we hypothesize that the wind stress curl (WSC) may affect recruitment through several mechanisms, including Ekman pumping/suction of deep nutrient-rich water to the surface, meridional transports, fronts position, and through lagged effect on basin-scale oceanographic properties. In particular, the long-term mean location of the transition zone between areas having positive and negative WSC (the WSC zero-line) coincides with the location of the largest BW spawning area in the Northeast Atlantic. Consequently, WSC variability in this region could potentially affect BW recruitment, through changes in upwelling/downwelling intensities, vertical mixing, and lateral transport processes.

We assess the relationship between WSC variability in the zero-line region and a BW recruit survival index between 1980 and 2021. We found that coupling stock-recruitment relationships to local indices of WSC variability near the zero-line significantly increased explanatory power (up to ~50%), especially if the recruit index was lagged 1 year behind the WSC variations. Using WSC as a driver of BW survival greatly improves the prediction abilities and forecast horizon of BW recruitment. It also suggests new processes driving BW survival which can be mechanistically investigated in the future, and could potentially inform sustainable, ecosystem-based management practices for this important fishery resource.



Ichthyoplankton communities at the northern Canary Current Upwelling Ecosystem - interannual variability and environmental forcing

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The western Iberian continental shelf is a highly dynamic and productive area with a strong latitudinal gradient, located in the northern part of the Canary Current upwelling system (North Atlantic Ocean). This area has a critical socio-economic role, as it supports a wide range of fishing activities. Despite its importance, few studies have focused on the ichthyoplankton communities. In this study, we analyzed for the first time the larval fish communities along the entire Portuguese coast, during the springs of 2018 and 2019. The aim was to assess the interannual variability in spatial distribution of larval fish communities and their relationship with dominant environmental conditions. A large diversity of species was identified, with the most representative families being Clupeidae, Gobiidae, Callionymidae, Sparidae and Blenniidae. *Sardina pilchardus* and *Engraulis encrasicolus* were the most abundant species, and a higher species diversity was observed in 2019. Our results show a latitudinal trend in the structure of ichthyoplankton communities, with specific taxa (e.g. Sparidae) prevailing in the southernmost areas, which reflected the dominant sea surface temperature gradient, the main driver of species distribution in the study area. This study provides key information that can be used to detect future changes in the fish communities of the western Iberian coast, and highlights the need for comprehensive monitoring programmes for effective management and conservation of fish and marine ecosystems.



Larval swimming and orientation behavior of the amphidromous goby, *Sicydium* spp., of São Tomé Island

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The *Sicydium* spp. of São Tomé and Príncipe (STP), LittleFish or ‘peixinho’ are amphidromous gobies that inhabit rivers with waterfalls and require freshwater-marine connectivity in their early life history. Adults spawn in freshwater and newly hatched embryos drift seawards; pre-settlement larvae must migrate back to the watershed and climb the waterfall into the freshwater stream where they mature and reproduce. While they are endemic of STP, their larval ecology and settlement migration are unknown. Recent studies have shown clear evidence of sustained swimming and orientation for most of pre-settlement larval stages of fishes. Here we posit that infrasound generated by waterfalls may be perceived by peixinho larvae as a directional signal to return to the adult habitat. To test this orientation hypothesis, we collected pre-settlement larvae as they ingress to the watershed of Cascata de Praia Pesqueira. The morning of their capture, before metamorphosis, they were readily deployed at sea in a Drifting In Situ Chamber designed to quantify their orientation behavior. The tested larvae between January 13-18, 2023 were preserved for further morphometric and genetic studies. Our results reveal orientation ability that appears to enable them to recruit in large numbers to the watershed, following general principles of SAAB Theory. Yet their interactions with the waterfall soundscape are still far from being understood. This research will feed the open-source Connectivity Modeling System with swimming and orientation data to estimate fish population connectivity between the islands of STP, relevant for fishery and conservation management.



Life in the fast lane: revisiting the fast growth – high survival paradigm during the early life stages of fishes

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Early life survival is critical to successful replenishment of fish populations, and hypotheses developed under the Growth-Survival Paradigm (GSP) have guided investigations of controlling processes. The GSP postulates that recruitment depends on growth and mortality rates during early life stages, as well as their duration, after which the mortality declines substantially. The GSP predicts a shift in the frequency distribution of growth histories with age towards faster growth rates relative to the initial population because slow-growing individuals are subject to high mortality (via starvation and predation). However, mortality data compiled from 387 cases published in 153 studies (1971-2022) showed that the GSP was only supported in 56% of cases. We argue that the GSP has hindered progress in understanding recruitment dynamics of fish by discouraging a more holistic exploration of processes that underlie growth-dependent selection and subsequent recruitment variability, possibly owing to a publication bias. Stochastic simulations allowed us to assess the influence of key intrinsic and extrinsic factors on the characteristics of surviving larvae and identify knowledge gaps on the drivers of variability in growth-selective survival. We suggest caution when interpreting patterns of growth selection because changes in variance and autocorrelation of individual growth rates among cohorts can invalidate fundamental GSP assumptions. We argue that breakthroughs in recruitment research require a comprehensive, population-specific characterization of the role of predation and intrinsic factors in driving variability in the distribution and autocorrelation of larval growth rates, and of the life stage corresponding to the endpoint of pre-recruited life.



Influence of temperature on density and growth of capelin (*Mallotus villosus*) larvae around the Icelandic coast

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Recruitment success is dependent on sufficient number of eggs and larvae surviving these early life stages. Environmental factors, such as temperature and the presence of suitable currents to transport the larvae to favorable nursery areas, can affect survival. Capelin is an important forage fish found in the north Atlantic. The Iceland-East Greenland-Jan Mayen capelin stock spawns primarily in the south and southwest coast of Iceland. The larvae normally drift with the currents towards the nursery grounds north of Iceland, but the drift pattern has been shifting with a portion of the larvae drifting to the east coast of Greenland. Our objective was to understand the influence of environmental factors on the distribution, density, and growth of capelin larvae in Icelandic waters. This could potentially lead to a better understanding of the reasons behind variations in recruitment and improve the management of this stock. Larvae were collected around the Icelandic shelf in May 2017-2020. Our results showed large density of larvae west of Iceland, and that warmer waters ($>7.5^{\circ}\text{C}$) had a negative effect on capelin larvae density. Overall, larvae were predominantly small ($<20\text{mm}$) over the years. Some large larvae ($>20\text{mm}$) were found north, northwest, and east of Iceland. Additional sampling was done in August and capelin larvae were found in lower density, with length a range of 20 to 40mm. Small larvae ($<20\text{mm}$) were mainly found in the south and southwest areas. In-depth growth studies are in progress to provide indication of slow-growing or a second wave of capelin spawners.



Spawning habitats created to promote recruitment of a key forage fish species in a large boreal reservoir

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Lake Saint-Jean is a large boreal reservoir of 1.053 square kilometers renowned for its recreational fishery of landlocked Atlantic salmon and walleye. Game fish productivity is directly related to the abundance of rainbow smelt, a dominant forage fish species whose recruitment may fluctuate by several orders of magnitude from year to year. The poor quality of sandy spawning habitats has been identified as a main factor limiting egg survival and larval production of rainbow smelt. To correct this situation, 25 rocky spawning habitats (total area of 3.800 square meters) were constructed in 2017 in known area for reproduction. The objective of this study is to verify the effect of the new spawning habitats on the deposition of rainbow smelt eggs, the abundance of larvae and, ultimately, the quality of the recreative catches in lake Saint-Jean. Egg deposition and larval abundance were measured before the establishment of spawning habitats and on three occasions after the construction (2017, 2019 and 2021). Results show that the new spawning habitats are utilized by rainbow smelt during reproduction. The abundance of larvae has increased. The effect of the new spawning habitats on the recruitment of the rainbow smelt population and on the production of game fish species is under watchful observation.



Improving Larvae Recruitment Survey Indices: A case study of North Sea Herring

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Recruitment is one of the major drivers of fish stock dynamics, and a robust index, which tracks its annual variability, is essential for fisheries management. Obtaining reliable estimates of future recruitment is still elusive, even if there are dedicated surveys for larvae or juveniles. In the case of North Sea herring (*Clupea harengus*) there is an early larvae (IHLS) and a late larvae (0-winter ring) survey (IBTS-MIK). The first providing Spawning Stock Biomass and the latter recruitment indices. While the IBTS-MIK index was considered a very good predictor of herring recruitment, its value has degraded recently with respect to forecasting. The IBTS-MIK takes place in January/February covering only the autumn spawning herring stock components, while the winter spawning component is neglected. With the winter spawning component now contributing a significant proportion of the stock, the MIK index became unable to represent the entire North Sea stock, while all other surveys, including the IHLS, do. In order to remedy this mismatch, a new survey timeseries that samples the offspring of the winter spawning component later, in April has been started. This survey has been carried out annually since 2018. With the time series available a new recruitment index is proposed, combining the standard MIK with the later survey, to provide a more reliable recruitment index for the whole stock.



S6. Application of “omics” in larval research

A proof-of-concept study to use DNA metabarcoding as a tool for species identification in ichthyoplankton samples

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Ichthyoplankton monitoring is important for stock assessments because fish eggs and larvae provide useful information on spawning grounds and seasons, allow the estimation of recruitment and spawning stock size, and identify changes in regional ichthyofauna. Usually, species identifications in ichthyoplankton samples are made by morphological inspection of the organisms, which requires taxonomical specialists who are frequently unable to guarantee accurate identifications due to the high similarity of characteristics between species. DNA metabarcoding is emerging as a cost and time-efficient complement that overcome these limitations. Species-level identifications on bulk samples and increased sampling frequency are made possible by this method, which combines DNA barcoding with high-throughput sequencing.

In the scope of the FCT-funded project “A-FISH-DNA-SCAN”, we conducted a proof-of-concept study to evaluate the efficiency of DNA metabarcoding in the identification of fish species in ichthyoplankton, in comparison with the morphology-based assessment. The samples were collected off southern Portugal last summer, on board IPMA’s RV “Mário Ruivo” using a Bongo net (335µm mesh size), and then sub-divided and randomly fixed with 96% ethanol or 4% formalin for molecular and morphological identification, respectively.

We were able to recognize several different species through morphological examination, but many were identified only at the family level. Comparatively, through the employment of three molecular markers (COI, 12S, and 16S), DNA metabarcoding, enabled the species-level identification of numerous eggs and larvae and resolved some of the uncertainties of the morphological assessment. Globally our results confirm the potential of metabarcoding to assess ichthyoplankton diversity and support fish stock assessments.



S7. Other contributed papers

The identification of flathead larvae (F. *Platycephalidae*) and their seasonal and spatial distribution off south-eastern Australia

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Platycephalidae is an iconic family of flathead fishes exploited by recreational and commercial fisheries around southern Australia. Despite this, species-specific characteristics of flathead reproduction such as larval distribution and seasonality remain largely unknown, as most flathead larvae could only be identified to family-level. Using a methodology integrating CO1 barcoding of morphotypes and subsequent detailing of morphological and pigmentation characters, 10 species of larval flatheads were identified in monthly ichthyoplankton samples collected between 2014–2021 at three oceanographic monitoring stations near Brisbane (27°S), Sydney (34°S) and Hobart (43°S) off eastern Australia. Seasonal water temperature significantly influenced the larval distributions, with distinct taxa characterizing the larval assemblages of each site. Species-specific larval abundances were analyzed with coincident oceanographic data to elucidate spawning seasonality and preferred environmental conditions for each taxa across latitudes. The paper will describe the temporal and spatial distribution of different species of flathead larvae across the three NRS sites based on adult distributions and reproductive patterns.



Ontogeny and morphology of the body axis of the Chilean Anchovy (*Engraulis ringens*)

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The Chilean/Peruvian anchovy, *Engraulis ringens*, is the one of the most productive and exploited wild-caught fish globally. Despite its extensive distribution from the central-south of Chile to Peru, many aspects of its biology, including skeletal development, remain poorly understood. In this study, we aimed to fill this knowledge gap by describing the appearance and transformations of elements of the axial skeleton in this species from small planktonic larvae to adults, using cleared and stained specimens for bone and cartilage. Our focus was on the positioning of fins along the body axis, and we found a significant anterior shift in fin placement, which we compared to the morphological changes of each fin. We also explored the regionalization of the anchovy body axis in relation to other teleosts. By providing a comprehensive morphological and developmental framework, our study may facilitate further natural history research, which is critical for the conservation and management of anchovies.



The use of macroalgae as an eco-innovative strategy to improve the immunity and antioxidant responses of juvenile farmed fish

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Aquaculture plays a key role in human nutrition and socio-economic development of many countries. However, disease outbreaks are a major bottleneck in the expansion of the sector, often being responsible for devastating animal and economic losses. Considering the projected abiotic changes due to climate change which are thought to facilitate the proliferation of certain pathogens and, at the same time, debilitate marine fish physiological status, the occurrence of disease outbreaks is expected to become more frequent and severe in the future. Overcoming this problem is certainly challenging, especially in the rearing of early-life stage animals that are usually more vulnerable to stress and exhibit higher mortality.

Currently, disease management in aquaculture often requires the use of pesticides and antibiotics, which raise major environmental and public health concerns. Hence, to avoid an intensification of such chemically-based approaches, the development of alternative eco-friendly strategies to manage diseases is urgently needed to assure aquaculture's sustainable growth in tomorrow's ocean.

The present study aimed to explore the use of macroalgae rich in immunostimulating bioactive compounds as an eco-innovative adaptation strategy to improve farmed marine fish immune and antioxidant mechanism. As such, two 60 days feeding trials with experimental aquafeeds containing different percentages of macroalgae (*Laminaria digitata* and *Asparagopsis taxiformis*) were performed using *Sparus aurata* and *Diplodus sargus* juveniles as models.

A multi-biomarker approach (combining histological, flow cytometry and biochemical methodologies) was carried out to determine the immunostimulating potential of the different functional diets.



Larval fish assemblage in Western coastal waters of Sri Lanka: Seasonal and spatial structure

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The study investigated the seasonal and spatial distribution patterns of larval fish in five off-shore and in-shore stations along West coast of Sri Lanka, which is an important area for the spawning and nursery of coastal fish. The samples were collected using vertical and horizontal WP2 hauls during different monsoon seasons. The results showed that Siganidae, Blenniidae, Clupeidae, Gobiidae, and Engraulidae families were the most dominant in off-shore sampling, while the Clupeidae, Engraulidae, Mullidae, Leiognathidae, and Pomacanthidae families were the most dominant in in-shore sampling. These families are ecologically and commercially significant, highlighting the importance of the area for pelagic and demersal fish as spawning ground. The species richness was found to be significantly different ($p < 0.05$) between the near-shore and off-shore locations, with the off-shore stations having higher richness. However, there was no significant difference in diversity ($p > 0.05$) or evenness ($p > 0.05$) between the two locations. In the near-shore stations, the mean abundance showed a decreasing trend from March to May for both vertical and horizontal samplings. The current study generates baseline data of the larval fish assemblage in near shore waters of Western coastal waters which provides useful information for sustainable fisheries management in the area. The results can be used to design effective conservation and management plans for the sustainable exploitation of the fishery resources in the region. The findings also emphasize the need for the continuous monitoring of larval fish communities in the area to ensure the long-term sustainability of the fishery resources.



Assessing the effects of nonylphenol and microplastics in *Sparus aurata* L., 1758 larvae: prey ingestion and dermal uptake pathways

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Understanding the risk posed on fish larvae upon microplastics (MPs) ingestion has been the main aim of several toxicological studies in the last years. However, since the tested concentrations frequently exceed environmentally relevant ones, the detrimental effects being reported are often unrepresentative of what occurs in nature. Moreover, considering the several contaminant pathways taking place in situ, laboratorial assays should include combined exposures in their experimental designs besides testing single stressors. Here we aimed to identify effects resulting from MPs ingestion (of polyethylene terephthalate fibers and ultra-high molecular weight polyethylene fragments) in fish larvae of *Sparus aurata*, which were simultaneously exposed to an organic compound (nonylphenol; NP), both by trophic transfer (through *Artemia nauplii*) and directly through dermal uptake, at environmental realistic concentrations. During a 6-day assay, 500 larvae were assigned to one treatment combining MPs and NP and to four control groups (absolute, solvent, NP, and MPs). Although no significant effects have been observed among groups, regarding mortality and biomarkers response, fish larvae simultaneously exposed to NP and MPs presented the lowest levels of antioxidant enzymes (catalase and glutathione S-transferase) and the highest levels of vitellogenin. Conversely, growth was significantly affected in larvae singly exposed to waterborne nonylphenol, suggesting that MPs ingested may reduce the NP body burden (detoxification role). Although the measured concentrations of NP in aquaria were considerably lower than the nominal concentration (5 µg/L), the tenuous effects here detected suggest harsh responses at higher NP concentrations (though still environmental realistic) and/or during a longer exposure.



Revising a lost knowledge: ichthyoplankton communities in the Israeli Mediterranean coast

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The current general knowledge regarding the diversity of the eastern Mediterranean ichthyoplankton is remarkably scarce, including its temporal and spatial distributions.

To overcome these knowledge gaps, we sampled neuston ichthyoplankton above the coastal, shelf and bathyal waters of the Eastern Mediterranean coasts of Israel, an oligotrophic basin prone to a continuous invasion of tropical biota.

To obtain species-level resolutions, we used DNA-based methods of molecular identifications for both single species and whole community levels.

Our results have uncovered the presence of 137 species which constitutes one third of the total local Osteichthyes fauna, evidencing a significant diversity of neuston ichthyoplankton. In addition, we showed that neuston communities can provide new knowledge regarding the presence of cryptic or rare species, and have a potential competence to reveal the presence of new non-indigenous species, by documenting fish larvae of three unreported species for the Mediterranean Sea.

Last, we posit that ichthyoplankton biomonitoring in surface water layer can complement any traditional fish biodiversity surveys throughout the marine environment.



Biogeography in the California Current Ecosystem from a Larval Fish Perspective

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Geographic features such as headlines, bathymetric features, and riverine input can create abrupt changes in ocean habitat characteristics. Depending on the life history of a marine species, geographical barriers can influence assemblage structure and patterns of diversity. In the California Current Ecosystem (CCE), which spans the coast of North America between Vancouver Island, Canada and Baja California, Mexico, multiple locations such as Point Conception and Punta Eugenia have previously been identified as geographic breaks for multiple marine species and populations. We explored spatial changes in the structure and diversity (number of species) of benthic versus pelagic fish assemblages at different geographic breaks by analyzing data of larvae systematically sampled across ~3000 km of the CCE during 2006 and 2008. For benthic fishes, distinct assemblages were found off two areas: Vancouver Island and Baja California. For pelagic fishes, distinct assemblages were found off five regions: Vancouver Island, Washington State to Point Conception, off southern California, off northern Baja California, and off southern Baja California. Latitudinal patterns in diversity also differed between benthic and pelagic species. The highest diversity for benthic fish was off Vancouver Island and there was no latitudinal trend throughout the rest of the CCE. For pelagic species, diversity increased equatorward from Vancouver Island to Cape Mendocino in northern California, remained stable between Cape Mendocino and Point Conception and then increased through southern Baja California. Results highlight the heterogeneity of marine life in the CCE and can help guide establishment of biologically-relevant conservation units.



POSTER CONTRIBUTIONS

S1. Global change effects on early life stages

PS1.1 Effect of temperature on routine behavior in early life stages of gilthead seabream, *Sparus aurata

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Since the industrial revolution, the intensification of the release of greenhouse gases has contributed to an increase in ocean temperature, particularly affecting ectotherm species that see their physiology and behavior changed. Such changes in behavior may arise through changes in routine swimming, foraging, boldness, among other fitness-related behaviors. However, the effects of temperature may be more complex than the simple relationship between stressor and individual performance, and may also affect intraspecific interactions, such as aggressiveness. The present study aims to evaluate the effects of temperature increase on boldness (assessed through time spent in refuge), routine swimming and aggressiveness (assessed through the number of chases and bites), in the early stages of Gilthead seabream (*Sparus aurata*), exposed for 3 weeks to the following temperature treatments: 19°C, 22 °C, 24°C (within the species thermal range) and 28°C (reflecting an extreme temperature treatment). Observations began as soon as the target temperatures were reached. For the quantification of behaviors, the focal observation method was used. Each observation lasted 2 minutes for the analysis of chase and bites frequency, followed by 2 minutes of observation for the quantification of routine swimming time, static in the water column and within the refuge. This procedure was repeated for 4 subjects per treatment, 3 times per week. Results suggest increased routine speed and aggressiveness with temperature, until 24°C, followed by decreased aggressiveness at 28°C. At this extreme temperature, fish did not use the available refuges.



***PS1.2 Vulnerabilities of Northwest Atlantic Living Marine Resources to Ocean Acidification**

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Ocean acidification (OA) poses a potential threat to marine fauna worldwide. It is anticipated that the Northwest Atlantic (NWA) will be particularly vulnerable to OA because atmospheric carbon dioxide (CO₂), the driver of OA, is absorbed more readily by cooler than warmer ocean surface water. Here we report on a comprehensive meta-analysis of the biological responses to elevated CO₂ in NWA marine macro-organisms. We consider NWA taxa as those found in waters from Cape Hatteras, North Carolina, USA to Newfoundland and Labrador, Canada. This project, funded by a joint USA-Canada OA partnership, had four objectives: 1) augment prior qualitative summaries of CO₂ effects on taxa from this region by including work conducted since the last qualitative regional review (Gledhill et al. 2015. *Oceanography* 28:182–197), 2) apply quantitative, hypothesis-testing analytical tools to these data, 3) create a public-facing infographic summarizing our results, and 4) support and train an early career scientist/student to lead the data collection effort. Data were collected from over 150 original studies. Response variables from the studies included calcification, growth, developmental, metabolic, and survival rates. For each study (species × experiment), we compared the change in response at ambient to CO₂ treatment levels using a log-transformed ratio. We then calculated the weighted average of response ratios to determine the cumulative effect size (+/- 95 percent confidence interval). Additional information on the focal species was used to construct testable hypotheses of the relationships between CO₂ sensitivities and the biological status of the subject.



PS1.3 Air exposure moderates effects of ocean acidification in early development of Pacific herring

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Ocean acidification can incur energetic costs of maintenance of acid-base homeostasis, leaving less energy available for growth and development in early life stages of fish. The Salish Sea is particularly impacted by ocean acidification, with CO₂ levels currently measured well above those predicted globally for the end of the century with climate change. However, for intertidally spawning fishes, such as Pacific herring, embryos are often air exposed for hours, potentially offering a refuge from high CO₂. We hypothesized that air exposure would be beneficial to the developing embryo due to a higher oxygen availability and permitting excess CO₂ to be off-gassed during emersion. To investigate the potential benefits of air exposure on CO₂ tolerance, we reared Pacific herring (*Clupea pallasii*) embryos under three tidal regimes fully crossed with three aquatic CO₂ levels and measured the effects on embryonic development and hatch, as well as carry-over effects on larval development and survival. Air exposure had significant positive effects on development, growth and survival, with some interactive effects with CO₂. Our research suggests that air exposure during low tide can be highly beneficial to intertidally spawning fishes and needs to be taken into account in climate change studies and future projections.



PS1.4 How different rearing temperature influence embryonic development and fatty acid utilisation of white seabream (*Diplodus sargus*) early life stages

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Embryonic and larval development of marine fish are strongly affected by environment temperature, due to its influence on the metabolism of poikilothermic organisms like fish. With this study it was aimed to gain insights on how temperature affects embryonic development and fatty acid mobilization pattern of *Diplodus sargus* early life stages. Eggs were incubated (4-cell stages) at 16°C (T16), 18 °C (reference temperature - T18) and 22°C (T22), in triplicate, and followed under the same conditions until mouth opening. Samples were collected at incubation (time 0; t0), at 12h (t12), at 24 h (t24) after t0, then at hatching and at mouth opening, to assess embryonic development (morula, blastula, gastrula, segmentation), biometric traits (egg size or larval length, yolk sac and oil droplet volume), total lipids content and fatty acid profile. Embryonic developmental stages were reached faster at higher temperatures. Total lipids content exhibited similar pattern of variation, decreasing during embryonic development to increase at newly hatched larvae, regardless the temperature. Temperature influenced how fatty acids were mobilized, resulting on differences at the different sampling points. Nonetheless at mouth opening higher content of polyunsaturated fatty acids (PUFA) were observed (e.g. ARA, EPA, DHA) among treatments, reflecting a similar strategy to preserve PUFA regardless the temperature. Still, at mouth opening fish larvae from T22 exhibited less endogenous reserves, which suggests a shorter period to established successfully exogenous feeding which may compromise survival success.



PS1.5 How extreme cold and warm oceanographic events influence larval fish assemblages in the southern region of the California Current off Mexico

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The fish larvae community in the southern region of the California Current (CC) was analyzed to test the hypothesis of a northward expansion of tropical species for the summer–fall seasons of La Niña (LN) 2010–2011, The Blob 2014 and El Niño (EN) 2015–2016. Interannual temperature anomalies (-5°C to 2°C), as well as a decrease in Chl-a (68%), and zooplankton density (71%), translated into drastic changes in the larval fish community, such as an unprecedented reduction of 82% in larval fish density previously unseen in the CC. The tropical species richness increased in the north by 46% and temperate species decreased by 65% in the south. The mesopelagic species richness increased in the north by 53%, as well as their relative abundance (92%). In the south, the species richness of the demersal component increased up to 39%, but they were co-dominant with mesopelagic species, accounting for 49% of the relative abundance vs. 47% of the demersal species. The magnitude of the changes in the community were unprecedented when compared with other warming events, such as EN 1983–1984 or EN 1997–1998. The differences were probably related to the presence of The Blob, which favored the transport of oceanic species into the neritic region of the CC region. In both cold and warm years, fronts and mesoscale eddies in the middle part of the Peninsula represented barriers to the latitudinal distribution of species, even during intense tropicalization processes since no latitudinal extensions in species distribution occurred.



***PS1.6 Immune responses of juvenile *Diplodus sargus* exposed to a marine heatwave**

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The aquaculture sector is a main contributor to global food security and economy, assuring the nutrition and livelihood of millions of people. Its contribution is expected continue increasing in the future, as to meet the needs of an also growing world population.

However, aquaculture's expansion faces many challenges related with the exacerbation of climate change effects, including the increased occurrence and severity of extreme weather events. In particular, marine heatwaves (during which seawater temperatures rise abruptly in a short time frame) are expected to have a preponderant role in farmed fish welfare and survival, especially in early-life stages (larvae and juveniles), by acutely exposing them to conditions outside their thresholds of physiological tolerance. Debilitated physiological status may likely compromise fish ability to deal with concomitant stressors, like disease outbreaks which, in turn, are also expected to occur more frequently and severely in a climate change context. Yet, little is still know about the effects of climate change-related stressors in farmed marine fish immune responses. This study aimed to unveil the impacts of a typical Mediterranean category II marine heatwave in the immune response of farmed juvenile *Diplodus sargus*. To this purpose, total erythrocytes and leukocytes counts and hematocrit (in blood), as well as, IgM content, antiprotease, peroxidase and lysozyme activities (in plasma) were determined. The outputs of this study will give insights to understand how resilient or vulnerable farmed juvenile fish are to these events, reinforcing the need to develop strategies towards climate change adaptation in the aquaculture sector.



PS1.7 Effect of temperature on European sardine larval growth, condition and survival throughout ontogeny

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The effect of temperature on sardine larvae growth and survival was tested during different times on the ontogeny of the larvae. We conducted experiments where sardine larvae 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 15 days post-hatching (dph), and survival was severely impaired at 19°C, tolerance to temperature increased sharply with age. When reared at optimal temperature during the 2 first weeks and changing temperatures at that time, survival rates were similar between 16 and 19°C, while growth rate increased with temperature for larvae reared until approximately 30 dph. Survival rate from 15 to 33 dph 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 the tolerance limits for temperature sharply increase throughout ontogeny.



S2. Larviculture

PS2.8 Identifying optimal conditions for early life-stage production of a cold-water wrasse *Tautoga onitis*: spawning, thermal regimes, culture density, and diet

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A first step in evaluating the aquaculture potential of a marine finfish is to identify the best methods for spawning adults and rearing the early life-stages of their offspring. One candidate aquaculture species in the NW Atlantic is Tautog (*Tautoga onitis*), a spring-spawning cold-water wrasse (Labridae) that occupies inshore benthic waters with a high density of bottom structure. The research team is composed of NOAA government scientists and support staff along with colleagues from the fish-farming industry. We are addressing three primary questions in our studies of tautog early life-stages that are designed to identify the optimal culture conditions for the production of juvenile tautog. First, how does egg quality vary within the spawning season, within the sequence of eggs released by individual females, and with whether eggs were obtained from spontaneous ripening or from accelerated ripening by hormonal injections? Second, what are the optimal thermal regimes during the embryonic and larval life-periods for production of hatchlings and juveniles? Third, how does the density and quality (enrichment) of live feed, and the stocking density of larvae impact juvenile tautog production? We are pursuing these questions with replicated factorial experiments under a controlled laboratory setting during the spring of 2023 and will summarize study results to date. The juveniles produced in these experiments and concurrent mass-rearing efforts will be used for larger tank grow-out studies on tautog juveniles by our collaborators at Ward Aquafarms.



PS2.9 Experimental and Breeding Centre at the NMFRI – new opportunities for the research on early life history of fish

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The Experimental and Breeding Centre of the NMFRI (Poland) enables experimental and breeding work in the field of biology and ecology of fish, including their early life stages. That covers species occurring in both fresh and saline waters. In the Center there are RAS circuits (Recirculating Aquaculture System) in several arrangements: hatchery, rearing and experimental working-area, with a total working volume of 40 m³. Individual circuits are equipped with mechanical filtration (microsieves), biological filtration (biofilters with a movable bed) and UV lamps, a system for generating and distributing oxygen, as well as a water cooling and heating system enabling operation in the temperature range from 5 to >25°C. Individual circuits, in addition to tap water, are supplied with an installation enabling the supply of water to the systems with various parameters, prepared in three separate tanks with vol. approx. 4 m³ each. The most important water parameters in individual circuits (oxygen concentration, temperature, salinity, pH) are monitored by the OxyGuard Pacific system and made available via the Internet on external computers and mobile devices. That includes alarm notifications about exceeding the assumed thresholds of individual water parameters. The operation of all devices is protected by an independent power generator. The first project conducted at the facility (PIKE, agreement no. 00002-6520.13-OR1100004/19; PO RYBY 2014-2020) is linked to pike *Esox lucius* rearing in different conditions (eggs incubation, larvae rearing) and large-scale fry production with acclimatization to saline waters for stocking coastal Baltic areas.



PS2.10 Optimizing feeding protocols for zebrafish larvae: effects of co-feeding with live prey and microdiets on growth and skeletal deformities

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Zebrafish larvae are widely used in biomedical and aquaculture research, but there is still no consensus among the zebrafish community on the most effective feeding protocol. In this study, we investigated the effects of different dietary conditions on zebrafish larvae growth, survival and skeletal deformities. We tested different feeding protocols: a rotifer-based and extended co-feeding regime with *Artemia* nauplii or rotifers, a microdiet-based feeding protocol, and an early transition to microdiets. Our results show that a rotifer-based and extended co-feeding regime with *Artemia* nauplii or rotifers resulted in decreased size dispersion and promoted larvae growth compared to the microdiet-based feeding regime or an early transition to microdiets. The larvae survival rate was also higher in the rotifer-based and co-feeding regimes. However, the use of a microdiet-based protocol reduced the incidence of skeletal deformities. These findings suggest that a better understanding of zebrafish larvae nutritional requirements is necessary to promote growth without compromising skeletal formation. The results also indicate that a rotifer-based and extended co-feeding regime with *Artemia* nauplii, or rotifers may be a more effective feeding protocol for zebrafish larvae. Altogether, more research is needed to understand the long-term effects of these protocols towards establishing a standardized feeding protocol for zebrafish larvae.



S3. Larval Trophic Ecology

PS3.11 Diet of Small Pelagic Fish larvae – insights from metabarcoding of larval stomach contents

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Small pelagic fish (SPF) have a key ecological role in their ecosystem and are important fisheries resources in many regions around the globe. A good understanding of the trophic ecology of these important fisheries resources, and especially at the early life stages, is essential information to allow predictions of recruitment strength based on the conditions experienced at the larval stage. While the diet of SPF is known for the adult stage of most species, little is known for larval stages. Mostly due to the low number of studies for most species and the high vacuity rate of SPF larvae based on traditional gut content analysis. Here we apply complementary molecular methods to study the diet of sardine and anchovy larvae collected from the Western and Southern Iberian coasts. Specifically, we performed metabarcoding of stomach contents from pooled larval samples, and compare it with results obtained by visual inspection of prey items. To maximize detection of the expected prey diversity, different molecular barcodes were used to target different taxonomic prey groups including prokaryotic and eukaryotic phytoplankton, and eukaryotic zooplankton. Application of metabarcoding to stomach content analysis of early life stages holds great potential to unravel the hidden prey diversity in the challenging small size scales of SPF larvae.



S5. Larval survival and recruitment variability

PS5.12 Do sardine and anchovy larvae share the same spots? *Sardina pilchardus* and *Engraulis encrasicolus* larvae distributions in NW Iberian waters.

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The most relevant small pelagic fish in the NW Iberian shores is typically sardine (*Sardina pilchardus*). However, its abundance is known to suffer (sometimes pronounced) fluctuations associated to environmental forcing and fisheries pressure. In recent years, while sardine attained one of the lowest densities recorded for the stock, a sudden increase in anchovy (*Engraulis encrasicolus*) was observed. In the present work, we investigate the co-existence of sardine and anchovy larvae during two periods, separated by a decade, first, when sardine was clearly dominant and then, when anchovy numbers peaked. The observations were conducted, in 2008 and 2018, during the regular acoustics surveys carried out by IPMA every spring. The high spatial resolution of the Continuous Underway Fish Egg Sampler (CUFES) results granted detailed mapping of the larvae and environmental variables distributions. The spatial analyses of the larval phase of the two species are discussed in view of the adults distribution and their spawning areas and also with regard to the recruitment grounds.



***PS5.13 Importance of growth history at first feeding stage for development and survival of larval yellow goosfish verified in laboratory experiment**

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Larval yellow goosfish *Lophius litulon* emerge from the gelatinous egg masses at the sea surface of coastal region. Larvae at the emergence stage were caught using plankton nets in coastal region, however, the subsequent larvae and juveniles were rarely collected. Therefore, environmental conditions of their nursery ground and mortality process have not yet been clarified. This study examined the effect of growth rate at first feeding stage on survival and development of the subsequent stages and appropriate environmental condition for the first feeding stage. Larvae from two egg masses were reared until ca. 30 days after hatching (DAH). Survival rate in the tank remained almost 100% during 8–15 DAH and then sharply decreased to 31% by 20 DAH. Width of the outer 5 daily increments from the 2nd check (formed during start of exogenous feeding) in lapillar otolith were measured to analyze their growth rate. Comparing the larvae that lived more than 25 days (SL) and the larvae that died by 20 DAH (DL), SL's increments were significantly wider than DL's. Number of dorsal fin rays of the larvae representing their developmental stage ranged 2–4 among SL samples. The larvae with 4 dorsal fin rays tended to have wider increments at first feeding stage than the larvae with fewer fin rays among SL. Our results suggest that faster growth at first feeding stage enhances survival and leads to superior development 1 month later. Therefore, the larvae at first feeding stage must distribute in areas with a high prey biomass.



***PS5.14 Larval retention and recruitment of small pelagic fish in a boreal fjord**

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Small pelagic fish play a key role in marine ecosystems, as they constitute essential links between secondary producers and higher trophic levels in food webs. These short-lived species experience highly fluctuating abundance levels, associated to variability in recruitment success. Fish populations that rely on estuaries as nursery areas generally experience larval drift and retention as major mechanisms regulating early life survival. In the Saguenay Fjord, Quebec, two osmerid species dominate the ichthyoplankton, the rainbow smelt (*Osmerus mordax*) and the capelin (*Mallotus villosus*), both representing important forage fish populations for the Saguenay—St. Lawrence ecosystem. They spawn during spring in the upper fjord and larvae drift downstream, subject to interannual variations of the Saguenay River discharge. The upper Saguenay Fjord comprises a deep wide inlet, Baie des Ha! Ha!, which acts as a retention zone for larval feeding, with relatively high abundances of *Eurytemora affinis*, a preferred copepod prey. It is hypothesized that in years with low discharge, osmerid larvae are more likely to drift into this inlet, where survival rates and recruitment potential are enhanced. In this study, we have investigated this retention mechanism, by studying the occurrence and abundance of osmerid larvae in June 2019, and how they related to zooplankton, hydrography and drift currents. Located in an inhabited region, the Saguenay Fjord ecosystem remains little studied. The investigation of small pelagic fish recruitment and its mechanisms will provide a basis for improving conservation strategies in this ecosystem, while generating fundamental knowledge on dynamics of fish early life stages.



PS5.15 Larval herring (*Clupea harengus* L.) abundance in the Pomeranian Bay (Baltic Sea) during 2007-2015 and 2019-2022

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The Pomeranian Bay basin is a convenient place as a spawning ground for many fish species, including herring, despite of the variability of its condition as sea waters and rivers mix here or high pressure of human activities. The intensity of herring spawning fluctuates in subsequent years, and regardless of its intensity, the degree of survival of early stages of development, i.e. the efficiency of reproduction, is also variable. Herring larval abundance surveys are all the more important as the spawning stock biomass of both the Western and Central Baltic stocks has declined from approximately 720.000 tonnes in 2011-2017 to 460.000 tonnes in 2021.

Sampling was carried out in spring, and it was found that herring larvae occurred in the Pomeranian Bay area between the second decade of April and the second decade of June, with a peak in abundance in late April to mid-May (in different years). In the last research season, the average number of herring larvae was only 13 pcs./100m³, which is the second worst multi-year result (compared to the years 2007-2015 and 2019-2021). It is still difficult to assess whether this is the effect of changes in environmental conditions in which, for example, eggs do not develop properly, or rather a decreasing spawning stock.



PS5.16 Effects of the vernal thermal bar on water quality, density and distribution of plankton and fish larvae in Lake Michigan, USA

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Thermal bars form in spring in nearshore waters of deep temperate lakes, and may structure horizontal distributions of temperature, nutrients, phytoplankton, zooplankton and larval fishes. Despite this, little is known of the effects of the thermal bar on distribution, density and growth of fish larvae, their spatial overlap with preferred temperatures and prey, and ultimately recruitment. In spring of 2015, 2018 and 2019, we sampled the environmental conditions and biota (including larval fish) inshore, offshore, and at the thermal bar from April to June. We used Niskin bottles to collect water for nutrient analysis and chlorophyll a, and nets to estimate zooplankton and fish larval densities. We tow-yo'd a laser optical plankton counter and MOCNESS to get continuous measures of temperature, light, chlorophyll, zooplankton and larval fish densities around the thermal bar. In 2015 and 2019, chlorophyll a and zooplankton biomass were significantly higher inshore than offshore of the thermal bar. Zooplankton biomass, nutrients and chlorophyll were retained inshore of the thermal bar in all years. In 2018 and 2019, larval fish densities peaked in mid-May, with deepwater sculpin *Myoxocephalus thompsonii*, burbot *Lota lota*, and yellow perch *Perca flavescens* dominating the catch. Highest densities of deepwater sculpin were found offshore of the thermal bar, whereas high burbot and yellow perch densities were found inshore and close to the thermal bar. Continued work will investigate how retention of zooplankton by the thermal bar may influence larval fish growth.



S6. Application of “omics” in larval research

***PS6.17 Using morphology and molecular techniques to describe fish larvae communities in west-African insular mangroves**

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The identification of fish larvae is extremely difficult in areas with limited information, such as the case of West African coastal waters. This limits the proper description of larval fish communities existing in important nursery areas such as mangroves. A study was conducted to identify the larval fish assemblages in two mangroves of São Tomé Island using morphological and molecular techniques, in an effort to contribute to increase our know-how of larval fish assemblages in poorly studied regions. The morphological identification of fish larvae was made up to the family level for 89.5% of the samples, while the remainder were identified to the genus (1%) or species level (9.5%). Larval gobiids and eleotrids were especially difficult to identify due to overlapping meristics and lacking available morphological descriptions. Consequentially, the specimens of these taxa were identified only to family level and separated on different morphotypes. Molecular techniques identified 48.6% of samples to the species level and 2.8% to the genus level while 48.6% had no match. However, 17.6% of larvae were molecularly mismatched at the species level. This study found six new species in their larval stage compared to previous works done on adult and juvenile fish in the same mangroves, being the first record on the fish larvae communities. The findings suggest that the mangroves of São Tomé Island serve as nursery areas for fish larvae, including species that do not occur on these systems in older life stages, supporting their importance at a local level for commercially important species.



PS6.18 Old meets new: 1. Morphological analyses of Guadiana ichthyoplankton as a support for DNA-based approaches

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Fish larvae and eggs have been conventionally identified through their morphological features and largely used in fisheries sciences for stock assessment and management. This knowledge has contributed to recognize seasons and spawning areas, estimate adult populations biomass, assess the spatial-temporal changes in composition and abundance of fish communities, and identify and evaluate new fisheries resources.

DNA-based methods for species identification have evolved in the last years. However, morphology-based taxonomic expertise is still necessary to guarantee accuracy and consistency among methodologies. This is the goal of the FCT-funded project “A-Fish-DNA-Scan”, that applies a traditional and a DNA-based approach to identify ichthyoplankton species from the Guadiana River estuary in the South of Portugal.

For one year, plankton tows (500µm mesh size) were monthly deployed in three different sampling points along the lower estuary. Samples were fixed and later sorted. The ichthyoplankton was counted and identified until the lowest taxonomic level possible. Environmental parameters such as temperature, salinity, chlorophyll a, and water clarity were also registered.

Through the conventional (morphology-based) methodology, more than 3500 eggs and 1100 fish larvae were found. Different taxa were identified, including 22 different species, 5 genus and 4 families. Some individuals were not identified, as well as most of the eggs. Overall, the most abundant species was *Sardina pilchardus*, but taxa as *Engraulis encrasicolus* and *Pomatoschistus* spp. also presented a high number of individuals. Autumn and winter showed lower abundances and species richness. These morphological analyses will now support DNA metabarcoding studies regarding the same ichthyoplankton communities.



PS6.19 Old meets new: 2. eDNA metabarcoding vs morphological analyses for detecting ichthyoplankton in the Guadiana estuary

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The availability of high-quality monitoring data for fish stock assessment is pivotal for the management of fisheries resources. DNA-based tools, namely environmental DNA (eDNA) metabarcoding, has been increasingly explored and adopted as a tool to identify marine species and shows great potential for biodiversity monitoring. However, its proper application still requires optimization and standardization, particularly on the markers and primers to be employed. eDNA fish community-derived data were compared to morphologically-based ichthyoplankton identifications. Two liters of water were collected at 3 sites along the Guadiana estuary, from April 2022 to June 2022, and eDNA was captured, extracted and amplicons analyzed by Illumina MiSeq. The efficiency of three different genetic markers (COI, 12S rRNA, 16S rRNA genes) and primers was tested to recover fish species diversity. We selected a primer cocktail and the primer-pair mlCOIintF/LoboR1 for COI; MiFish U-E for 12S; and Fish16sFD/16s2R for 16S. Preliminary results show that eDNA was able to consistently detect more genera than morphologically-based analyses. The large majority of the genera (> 60%) that were identified morphologically were also found in eDNA from water samples. All 3 markers combined detected 38 fish species, however, only 6 (16%) were simultaneously detected by all of them. Individually, 16S performed the best and was capable to identify 19 fish species, whereas 12S identified 17 species and COI just 14. Thus, our research corroborates the potential of eDNA metabarcoding as a tool for biomonitoring fish diversity. Yet, a combination of markers and primers should be used to increase species detection.



***PS6.20 First steps in exploring deep-sea fish larval development in the eastern Mediterranean Sea**

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Little is known about the biota of the deeper part of our oceans due to its inaccessible location and harsh conditions. This study aims to overtake these limitations by obtaining knowledge on the larval development of deep-sea fishes that utilize the upper surface layer for their dispersal and early life development. During December 2022, fish eggs were collected from the offshore surface layer of the Israeli Mediterranean Sea above bottoms of 1000-1300 m and using horizontal hauls of Manta net, 300 µm mesh.

The fish eggs were taken to the laboratory and were documented and measured every nine hours. Images were used to create an ongoing database, and growth rate comparisons were made for the different specimens. After mortality, each egg's DNA was extracted separately, and its barcoding gene (COI) was amplified to obtain species-level taxonomic identification.

The final results were based on 17 of the 29 collected fish eggs with reliable matches of three species: *Brama brama* (11 eggs), *Polyprion americanus* (4 eggs), and *Chauliodus sloani* (2 eggs). This study's preliminary results regarding spawning seasonality, egg morphology, and lecithotrophic larval development of these three cryptic and rare species are undoubtedly novel.

This promising potential for investigating the early life history of deep-sea fishes will be further pursued during this study, with an overarching goal to improve our knowledge of this less-explored oceanic zone.



S7. Other contributed papers

PS7.21 Morphological description of *Pogonias courbina* (Lacepède, 1803) larvae

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Ichthyoplankton studies contribute to ecological analyses and management of fish stocks. However, during sampling collected fish larvae are difficult to observe and study so preserving individuals is essential and must be carried out as soon as possible. But fixation alters body proportions and causes the disappearance of pigments that are used to identify the organism. To fill this gap, studies of larval description with morphometric and meristic analysis are necessary. The focus of this study is the Sciaenidae *Pogonias courbina*, which lacks description of its initial life stages and studies on morphometric and meristic changes during development. Larvae collected during spawning in the Laboratory of Marine and Estuarine Pisciculture of the Marine Aquaculture Station (EMA-FURG) were anesthetized (Eugenol 30ppm), measured, illustrated and preserved in formalin 4%. Larvae were measured with a stereoscopic microscope and the use of ImageJ software, and were illustrated with a camera lucida. As a result of this study, we describe and illustrate the early stages of *P. courbina* from eggs to juvenile. The eggs collected were transparent, with non-segmented yolk. 3 days post-hatch (DPH), yolk was completely absorbed. Mouth and anus opened at different times, and the exogenous feeding started at 4 DPH. Flexion of notochord started at 9 DPH, and metamorphosis from larvae to juvenile was concluded in roughly 30 DPH. The study will fill the knowledge gap of the initial stages of *P. courbina* and also serves as a tool for use in biological collections, museums and population studies in management and preservation.



PS7.22 Zebrabloom: novel microalgae product for zebrafish nutrition

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Zebrafish is a model organism widely used in multiple scientific areas. However, the poor understanding of this species nutritional requirements leads to a lack of dietary protocol standardization among zebrafish facilities. Feeding zebrafish with live prey, such as rotifers enriched with microalgae (*Nannochloropsis* sp.) is commonly applied in zebrafish early life stages, since it improves survival due to their high digestibility. Zebrabloom project aimed to develop of a new microalgae diet nutritionally adequate for zebrafish larvae and broodstock (as well as rotifers culture) towards the improvement of breeding performance and larvae quality. A liquid concentrate combining different microalgae (*Nannochloropsis oceanica*, *Tisochrysis lutea* and *Tetraselmis chui*) was formulated as a multipurpose product for all zebrafish life stages. Zebrafish broodstocks (n=20 males and n=20 females per treatment) were fed during 4 weeks with: commercial microdiet (CM), rotifers enriched in Zebrabloom, Artemia nauplii (Art), Artemia metanauplii enriched in Zebrabloom (ArtZ), and rotifers enriched with two microalgae commercial products (CP1, CP2). Each broodstock was set to breed once a week (1:1 sex ratio). Broodstock fed with rotifers enriched with Zebrabloom produced the highest number of eggs, being significantly higher than those produced in CM, Art, ArtZ treatments. No significant differences were found in hatching rate between treatments. Samples from diets, rotifers and Artemia enriched with microalgae, laid eggs, larvae and adult fish were collected to evaluate the impact of the different nutritional profiles. Results suggest that Artemia is inadequate for zebrafish broodstock nutrition and rotifers enriched in Zebrabloom are beneficial for zebrafish nutrition.

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***PS7.23 Do early life stages of fish show signs of personality?**

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Most studies carried out on personality recognized that personality is defined by behavioral traits consistent through time and/or contexts. Considering that personality has a great influence on organisms' fitness, understanding the variation and flexibility of personality will have significant ecological implications. Recent studies have challenged the previously assumed stability of personality types across lifetimes, highlighting the need to understand better how repeatability (within life history stages) and consistency (between life history stages) differ throughout ontogeny.

In this study, we performed behavioral tests to characterize boldness (emergence test), exploration (open field test), activity (velocity swimming), and aggressiveness assays (mirror-image test) in larval stages of a temperate reef fish, the gilthead seabream (*Sparus aurata*). A total of 37 larvae fish, ranging from 1,926 to 1,933 cm, were tested twice in each assay.

The findings suggest that there is a lack of personality traits, such as boldness and exploration, during this phase, as none of the traits were replicable because of the limited inter-individual diversity in behavior. Behavioral flexibility is likely to be necessary for animals that undergo early life.



PS7.24 Comparison of the juvenile morphology and assessment of ontogenetic changes in morphology of three species of seahorses *Hippocampus* spp. from south-eastern Australia

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Seahorses are a charismatic component of the fish fauna in estuarine and coastal marine water. In addition to their unique morphology and reproductive strategy, they are threatened by habitat degradation. The taxonomy of seahorses has been problematic with many species misidentified in museum collections. This poster will describe the morphology of newly hatched juveniles and ontogenetic changes with growth of three species of seahorses, *Hippocampus whitei*, *H. abdominalis* and *H. breviceps* from south-eastern Australia. We will describe and compare differences in morphological characters of juveniles, such as the number and location of spines on the body and the structure of the coronet on the head, which are important features for identification of adults of different species. The ontogenetic changes in morphology of the juveniles will also be compared with the adult morphology of the three species.



PS7.25 Drivers of sardine larvae survival in the northern Canary upwelling system

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Southern Iberian sardine stock biomass decreased by 60% between 2006 and 2010, and in 2016 sardine recruitment reached the lowest level in 40 years. As a contribution to the understanding of the causes behind this long period of low annual recruitments, we analysed the growth and nutritional condition of wild sardine larvae collected at the beginning and end of the spawning season within Atlantic Iberian waters, located at the northern Canary upwelling ecosystem. Larvae were collected with Bongo nets during autumn and spring acoustic surveys. To study larval growth, individual larvae were measured and aged by daily increment counts in the otoliths. The total and recent otolith growth was also determined. The condition of individual larvae was assessed by the DNA per DW in the muscle. Growth and larvae condition were modelled as a function of several environmental parameters to investigate the main drivers for larvae survival in the wild.



PS7.26 Ichthyoplankton spatial variability in the SW Atlantic during the relaxation phase of coastal upwellings

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We investigated the latitudinal and continental shelf–slope gradients in ichthyoplankton structure and oceanographic features in the Campos Basin during the relaxation phase of coastal upwellings. This region, located on the continental margin of SE Brazil, is the most productive offshore oil basin in the country and has ecological significance due to the presence of areas with mesoscale processes generated by the interaction of continental shelf and slope circulation with deep water masses. The present study registered 3892 fish eggs and 10 030 larvae from 36 sampling points, with an average of 22 eggs and 56 larvae per 100 m³. This totaled 250 taxa (5 for fish eggs and 248 for larvae), divided into 80 families and 145 genera. The species distribution demonstrated significant geographical heterogeneity, mostly linked to hydrological features. Greater densities were often related to areas with higher nutrient concentrations. Furthermore, when discriminating species distribution patterns were investigated using distance-based redundancy analysis, certain species were found to be related to colder waters on the continental shelf. Thus, it is assumed that the Campos Basin ichthyoplankton assemblages use eddies and meanders from the Brazil Current as a transport or accumulation mechanism to nutrient-rich areas for the development of this early life stage over the autumn–winter period. Overall, this study contributes to a better understanding of the mechanisms underlying spatial heterogeneity in ichthyoplankton communities in the SW Atlantic.



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