

5th INTERNATIONAL OTOLITH SYMPOSIUM

October 20-24, 2014



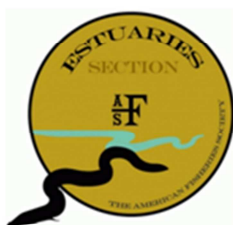
**Casal de Peguera, Peguera (Calvià),
Mallorca, Spain**

An



Science Symposium

SPONSORS



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Beatriz Morales-Nin, IMEDEA (CSIC-UIB), Esporles, Illes Balears, Spain

Audrey J. Geffen, University of Bergen and Institute of Marine Research, Bergen, Norway

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Audrey Geffen (University of Bergen, NO)

Beatriz Morales-Nin (IMEDEA (CSIC/UIB), ESP)

Welcome!

It is a great pleasure to welcome you to the 5th International Otolith Symposium, a continuation of the series started in 1993, to encourage the exchange of information and expertise and promote the development of new techniques and applications for otolith-based analysis in ecology, management, and conservation.

We are excited about the possibilities of this international forum to be the centre for the exchange of information on developments in the field, new techniques of analysis, statistical treatment, and indeed new areas of application. The aim of the Symposium is to explore the **use of calcified tissues as tools to support management** and define indicators at environmental, community, population and individual levels.

Our programme combines scientific presentations and workshops together with cultural and social events. We are proud of the high standard and number of presentations, and we hope that the recreational opportunities will encourage productive scientific discussions over food and wine!

We wish you a successful participation with hard work and fun!

We hope that your stay in the beautiful Island of Mallorca is pleasant.

Beatriz Morales-Nin & Audrey J.Geffen

Keynote Speakers

Theme I: Environmental indicators

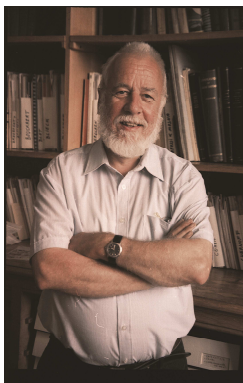
Invited speaker - **Rita P. Vasconcelos** Researcher, Centro de Oceanografia - Faculdade de Ciências, Universidade de Lisboa (Lisboa, Portugal) and Pôle Halieutique - Agrocampus Ouest (Rennes, France).



One of Vasconcelos' main research areas is centered on population connectivity, chiefly in measuring nursery function of fish populations via multiple tools. During her PhD, completed in 2009, she tested the match between the potential contribution of estuarine nurseries and their effective contribution to adult populations (using a set of commercially exploited fish species). More recently she has addressed connectivity of coastal populations of several species, through the integration of several natural markers. She is also a member of the ICES Working Group on the Value of Coastal Habitats for Exploited Species, in which population dynamics models are being applied to assess the value of coastal habitats for several species. Another core research area focuses on the development of modeling of fish populations and communities in estuaries with emphasis on biodiversity and ecosystem functions and services. In this context, research is directed towards enhancing predictions obtained with models, to forecast changes in fish populations and communities in face of natural and anthropogenic environmental change.

Theme II: Community indicators

Invited speaker - **Dirk Nolf** Associated Professor, IRSNB (Institute Royal des Sciences Naturelles) (Brussels, Belgium)



In the last twenty years, Nolf's research has been concerned with external morphology of fish otoliths, with special emphasis on the otolith-based reconstruction of fossil fish faunas. Another aspect concerns the use of otoliths for the identification of recent fishes and the interpretation of interrelationships between recent and fossil fish taxa. Presently, he is completing a book on the otoliths of recent fishes from the North Sea and the English Channel, in which otoliths of almost all species will be illustrated with an extensive series of their variability.

Theme III: Population indicators

Invited speaker - **Simon Thorrold** Senior Scientist, Biology Department, Woods Hole Oceanographic Institution (Woods Hole, USA).



Thorrold's work focuses on measuring animal movements in ocean ecosystems using a variety of tagging methodologies. Much of this research uses an eco-geochemistry approach - the application of isotope and trace element geochemistry to fundamental questions in population and community ecology. Independent approaches, including DNA parentage and electronic tagging, are also used to both verify results from the geochemical analyses and to generate and test new hypotheses on animal movements. This information on the influence of movements on population dynamics provides decision makers with a scientific basis for the conservation and sustainable management of ocean ecosystems.

Theme IV: Individual indicators

Invited speaker - **Peter Grønkjær** Associate Professor, Marine and Fisheries Ecology, Department of Bioscience, Aarhus University (Aarhus, Denmark).



Grønkjær's interest in how the environment impacts the ecology of fishes has led to research in topics ranging from the ecophysiology of zebra fish larvae to winter-migrations of cyprinids. However, the use of otoliths as proxies for growth, metabolism and size has been common to most of his projects, where they have been used to reveal size-selective mortality, links between otolith fluctuating asymmetry and nutritional condition, and selection against fish larvae with high metabolic requirements in food limited environments. Recently, the focus has been on the microchemistry otoliths, and, especially the potential use of stable carbon and nitrogen isotopes from the organic otolith matrix to explore food web structure and the trophic position of fish populations. The large collections of otoliths sampled over the last 100 years offer a unique possibility to explore changes in aquatic food webs and their effects on fish population dynamics.

General Programme:

The Symposium runs from Monday to Friday, with Wednesday dedicated to the parallel Workshops. The venue is the Casal de Peguera, a cultural center run by the Calvià City Council, located in Peguera.

For more information: <http://ices.dk/news-and-events/symposia/otolith/Pages/Venue.aspx>

Registration starts on Sunday, Oct 19th, at 18 h in the Palmira Beach Hotel (Peguera, <http://www.palmirahotels.com/en/beach.php>) continues Monday morning at the Casal venue.

Each day will be dedicated to one of the themes of the Symposium and will begin with a morning plenary session introduced by the keynote speaker, followed by oral presentations. In the afternoons, oral presentations will continue in two parallel sessions, followed by a round of mini-talks, "Otochuki" sessions, which close the day's work.

Poster presentations will be displayed throughout the symposium, with a special poster session on Tuesday evening. During the week a slide-show of selected communications will run continuously.

Support office: During the Symposium the registration and help desk will be open at the Casal.

Transfers: Transport will be provided from Palmira Beach Hotel to the Casal de Peguera at 8.30 every morning and back to the Hotel at the evening end of the programme. Transport will be arranged for people with lunch bookings at the Palmira Beach Hotel .

Awards: The best student oral and poster presentations will be rewarded with a prize provided by the Estuaries Section and the Early Life History Section of the American Fisheries Society.

Cultural event: The **Protolithe** installation is an Art and Science collaborative initiative between a poet, an analytical chemist, a fish ecologist and a music composer, based on the information retrieved from fish otoliths. **Protolithe** is a collection of poems, haïkus, questioning the epic journeys of migratory fish. The exhibit will be open during the symposium, upstairs in the Casal.

Social events:

Besides the morning and afternoon breaks, an **ice-breaker** with food and drinks will be held on Sunday Oct 19th at the Palmira Beach Hotel from 19 to 21 h.

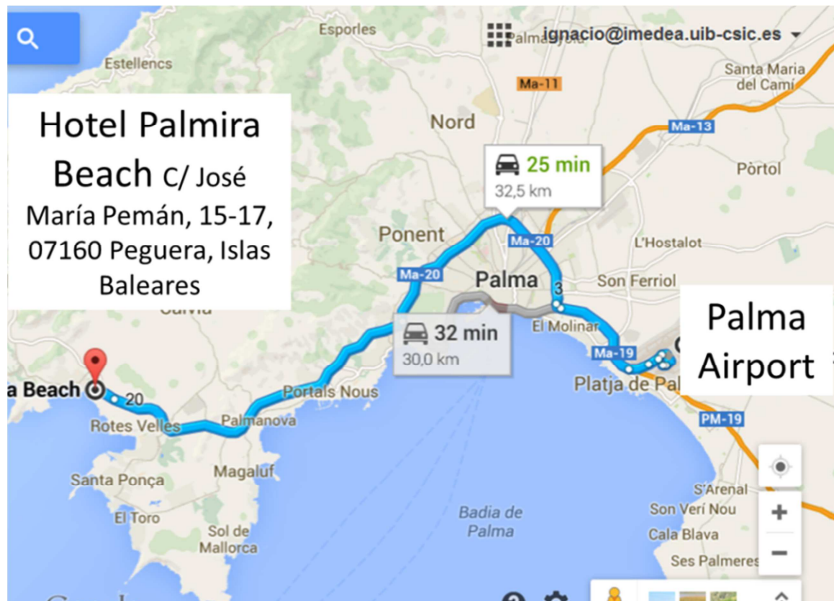
On Tuesday Oct 21st a **Poster Session** will be held with a Mallorcan wine and food tasting at the Casal de Peguera.

On Friday Oct 24th after the closing ceremony, a special "Demonic Mallorcan" **fiesta** will be held at the church square - 50 m from the Casal de Peguera. It is advisable to wear cotton clothing, long sleeves (such as the symposium T-shirt!) and a hat for active participation. Otherwise, watch the spectacle in safety from a short distance away. The evening social activities will culminate with a **Gala dinner**, a flamenco spectacle, and music at the Palmira Beach Hotel.

Palma Aquarium will offer special prices and offers to visit this extraordinary aquarium for a two week period encompassing the IOS dates (<http://palmaaquarium.com/>). See leaflet and entrance voucher in the symposium bag.

Mallorca has pristine rural areas and spectacular scenery in the north (Tramuntana coast). The city of Palma de Mallorca is full of interesting architecture and museums, including the imposing gothic cathedral and the old town. The Palmira Hotels will be happy to organize excursions for anyone interested in exploring

Location



Location of Casal de Peguera:



The Protolithe Installation

The Protolithe installation is an Art and Science collaborative initiative between a poet, an analytical chemist, a fish ecologist and a music composer, based on the information retrieved from fish otoliths. Protolithe is a collection of poems, haïkus, questioning the epic journeys of migratory fish. The installation is based on the results and tools used for collaborative research projects between ecology of diadromous fish and laser ablation analytical chemistry. Protolithe is composed of two parts. The first illustrates the geographic milestones of migrations, embodied by water steles, made of glass tubes filled with water from the different habitats encountered by the fish and engraved with the collection of poems protoscape. The second part of the installation is Odyssey, poems engraved in microscopic letters on otoliths, after the story of each individual fish. The installation is completed with a video of fish passage on the background and an immersive music. The installation was inspired by the fish individual migratory patterns retrieved from otolith microchemistry of the presented otoliths, formerly used in different collaborative projects addressing European eel, flounder and Shad individual diversity of migration pathways. Challenging laser ablation techniques were performed to both analyse specific life stages of fish, and engrave poems in 30 µm height letters.

Protolithe will be shown in the upper floor of the Casal de Peguera from Monday afternoon to Friday.

Protolithe exhibition sponsors:



Casal de Peguera meeting rooms



- A: Auditorium**
- B: Seminar Room**
- PC: Poster and coffee break area**
- R: Registration**
- Pro: Protolithe exhibition**

Programme Overview

Sunday Oct 19 th	Palmira Beach Hotel	Thursday Oct 23 rd	Casal
18:00	Registration	9:00	Sessions start
19:00-21:00	Icebreaker	10:45-11:15	Coffee Break
Monday Oct 20 th	Casal	13:10-14:30	Lunch
8:30	Registration	16:10-16:30	Coffee Break
9:30	Sessions start	19:30	Sessions end
10:45-11:15	Coffee Break	Friday Oct 24 th	Casal
13:10-14:30	Lunch	9:00	Sessions start
16:10-16:30	Coffee Break	10:45-11:15	Coffee Break
19:30	Sessions end	13:10-14:30	Lunch
Tuesday Oct 21 st	Casal	16:10-16:30	Coffee Break
9:00	Sessions start	19:15	Closing Ceremony
10:45-11:15	Coffee Break	19:40	Demon Spectacle
13:10-14:30	Lunch		Palmira Beach Hotel
16:45-19:00	Poster session	21:00-24:00	Gala Dinner
19:30	Sessions end		
Wednesday Oct 22 nd	Casal		
9:00	Workshops start		
10:40-11:00	Coffee Break		
13:00-14:00	Lunch		
15:45-16:00	Coffee Break		
18:00	Workshops end		

Programme

Sunday, 19 October				
REGISTRATION		18:00	Palmira Beach Hotel	
"A word in your ear(stone)" ICEBREAKER		19:00		
Monday, 20 October				
REGISTRATION		8:30	Casal saloon	
SYMPOSIUM OPENING		9:30	Auditorium	
Keynote Theme I: Environmental indicators		10:00	Auditorium	IA_Vasconcelos_Key (A.Geffen speaker presentation)
Coffee Break & Burn		10:45	Poster Room	
MORNING PLENARY SESSION		IA	10:00 - 13:10	Auditorium Moderators S.Campana, F.Cerna
Protoliths: protoscape and fish odyssey			11:15	IA_Daverat_00
Using fish ear bones as proxies for tracing changes in climate			11:30	IA_Gillanders_01
Investigating the bomb radiocarbon flux in the southern Pacific Ocean with otolith radiocarbon			11:50	IA_Grammer_02
Virtual beam shaping femtosecond laser ablation: a new tool for sensitive high spatial resolution otolith microchemistry.			12:10	IA_Pecheyran_03
Natural habitat contributes more to an estuarine fish production than artificial habitat does: variability in growth and mortality rates of larval and juvenile temperate bass cohorts estimated from otolith microstructures			12:30	IA_Shoji_04
Do variations in the elemental concentrations in otoliths of marine fishes really track variations in water chemistry?			12:50	IA_Trueeman_05
Lunch lapilli			13:10	
AFTERNOON PARALLEL SESSION		IB	14:30 - 16:10	Auditorium Moderators R.Vasconcelos, I.Catalan
Shades of Sophie Dove? The potential of eye lens chemistry as a complementary archive of environmentally relevant information			14:30	IB_Limburg_01
			14:50	
Ongoing ageing of a habitat-forming deep-sea coral <i>Solenosmilia variabilis</i> – information to assess recovery potential from anthropogenic impacts			15:10	IB_Tracey_03
Effects of ocean acidification on otolith growth and size-selective mortality of larval Atlantic cod and Atlantic herring			15:30	IB_Clemmesen_04
Detecting interannual shifts in otolith microchemistry before, during, and after the Deepwater Horizon oil spill			15:50	IB_Granneman_05
AFTERNOON PARALLEL SESSION		IC	14:30 - 16:10	Seminar Room Moderators A.Andrews, P.Grønkjær
A stitch in time: using chemical chronologies from otoliths to reconstruct larval dispersal environments			14:30	IC_Sweaver_01
Otolith-derived estimates of marine temperature use by West Greenland Atlantic salmon (<i>Salmo salar</i>)			14:50	IC_Power_02

A 200 year archeozoological analysis of Pacific cod life history as revealed through Ion Microprobe oxygen isotope ratios in otoliths 15:10 IC_Helser_03

Alaskan lake trout biochronologies as long-term indicators of climate and productivity 15:30 IC_Wilhelm_04

Insights into historical New Zealand marine shelf productivity using ancient fish otoliths 15:50 IC_Neil_05

Coffee Increments 16:10 Poster Room

AFTERNOON PARALLEL SESSION IB 16:40 – 18:00 Auditorium Moderators B.Gillanders, H.de Pontual

An investigation of connectivity among estuarine tapertail anchovies *Coilia nasus* from the Yangtze River, Yellow Sea and Poyang Lake 16:40 IB_Yang_06

Extracting environmental histories from sclerochronological structures - Recursive partitioning as a mean to explore multielemental composition of fish otolith 17:00 IB_Vignon_07

Relative contribution of estuarine nursery areas to the adult population of the temperate seabass *Lateolabrax japonicus* in Tango Bay, as revealed by otolith Sr:Ca ratios 17:20 IB_Yamashita_08

Near-reef elemental signals in the otoliths of *Pomacentrus amboinensis* (Pomacentridae) 17:40 IB_Sih_09

AFTERNOON PARALLEL SESSION IC 16:40 – 18:00 Seminar Room Moderators D.Tracey, C.Trueman

Contrasting environmental drivers of adult and juvenile growth in a marine fish 16:40 IC_Ong_06

Examining the interactions of growth, climate and recruitment of boarfish (*Capros aper*) for a better understanding of the recent population expansion 17:00 IC_Davies Coad_07

Broad and local scale drivers of growth of an estuarine fish species and implications for climate change 17:20 IC_Doubleday_08

Comparison of Slimy Sculpin (*Cottus cognatus*) annual growth in contrasting regulated and unregulated riverine environments 17:40 IC_Kelly_09

OTOCHUKI ID 18:15 - 19:15 Auditorium Moderator A.Geffen

Migrating Atlantic cod otoliths reflect movement through water masses: linking data storage tag data with high resolution trace element and isotope geochemical signatures 18:15 ID_Neville_01

Settlement variability of *Mullus surmuletus* by means of otolith sclerochronology 18:20 ID_Matteo_03

Growth of King George whiting: an investigation of climatic influences on otolith growth 18:25 ID_Mazloumi_05

Otolith chemistry discriminates water mass occupancy of Arctic fishes in the Chukchi Sea 18:30 ID_Norcross_06

Otolith biochronologies and the present, past, and future of the California Current upwelling system 18:35 ID_Black_07

Life-history connectivity and climate-related change in distribution of red hake (<i>Urophycis chuss</i>) on the northeast United States continental shelf.	18:40	ID_Robillard_08
Temperature and oxygen isotope variability of North Sea Basin shelf waters during the early Eocene, recorded by fish otoliths	18:45	ID_Vanhove_09
Does Ba:Ca and Sr:Ca ratios always reflect ambient water composition where Atlantic salmon (<i>Salmo salar</i>) inhabits?	18:50	ID_Bareille_10
Comments on the microstructure of vertebrae, and estimates of age and growth of the blue shark, <i>Prionace glauca</i> (L., 1758), in the southwest Atlantic	18:55	ID_Montealegre- Quijano_12

Tuesday, 21 October				
MORNING PLENARY SESSION	IIA	9:00 - 12:35	Auditorium	Moderators Y.Yamashita, B.Walther
Keynote Theme II: Community indicators		9:00	Auditorium	IIA_Nolf_Key (A.Lombarte speaker presentation)
What do we know about otolith function in hearing? – Insights from cichlids and livebearing fish		9:45		IIA_Schulz-Mirbach_01
Stable carbon isotopes in marine fishes otoliths		10:05		IIA_Shores_02
Eleven years of the interactive AFORO (Shape analysis of otoliths) database web site (2003/2014)		10:25		IIA_Lombarte_03
Coffee check		10:45	Poster Room	
Multi-decadal biochronologies indicate species-specific responses to environmental change		11:15		IIA_Izzo_04
A 100-year assessment of biological change in SE Australian waters: novel insight using fish otoliths		11:35		IIA_Morrongiello_05
Prey composition analysis using otolith morphology: a case study of bigeye tuna <i>Thunnus obesus</i> in the western Indian Ocean		11:55		IIA_Chang_06
The use of otoliths to examine trophic interactions between California sea lions and artisanal fisheries on the west coast of the Baja California peninsula		12:15		IIA_Tobar_07
Lunch lobes		12:35		
AFTERNOON PARALLEL SESSION	IIB	14:00 - 15:40	Auditorium	Moderators D.Nolf, J.Panfili
May otolith morphology be used for measuring biodiversity of marine fish assemblages?		14:00		IIB_Tuset_01
Inter-population differences in otolith morphology are genetically encoded in <i>Aphanius fasciatus</i> (Cyprinodontiformes, killifishes)		14:20		IIB_Reichenbacher_02
Evolution of trisopterine Gadidae with otoliths		14:40		IIB_Gaemers_03
Variability in the Santa Barbara Basin Fish Assemblage in the Last Two Millennia Inferred from the Fossil Otolith Record		15:00		IIB_Jones_04
Fish otolith assemblages on the Recent NE Atlantic sea bottoms		15:20		IIB_Lin_05
AFTERNOON PARALLEL SESSION	IIC	14:00 - 15:40	Seminar Room	Moderators K.Limburg, A.Volpedo
Determining the foraging grounds of the Great Cormorant by otolith shape and chemistry		14:00		IIC_Oehm_01
The use of otolith microstructure for analyzing effects of ectoparasites on nearshore fish larvae		14:20		IIC_Landaeta_02
Functional diversity and behavioral responses to depth gradients revealed by otolith morphology in deep-sea fishes		14:40		IIC_Chung_03
Discriminating cryptic speciation using multivariate analysis of otolith morphometrics		15:00		IIC_Wakefield_04

Connectivity and fish population structure: perspectives from otolith geochemistry and genetic markers 15:20 IIIC_Reis-Santos_05

OTOCHUKI	IIID & IVD	15:50 - 16:40	Auditorium	Moderator B.Black
Movements of the North Atlantic albacore (<i>Thunnus alalunga</i>) revealed by otolith $\delta^{18}O$, $\delta^{13}C$, Sr:Ca, Mg:Ca, Mn:Ca and Ba:Ca chronologies		15:50		IIID_Fraile_01
Stock identification of <i>Sprattus sprattus</i> within the Celtic Sea Ecoregion using otolith shape and microstructure		15:55		IIID_Moore_02
Residency and Habitat Use of Southern Flounder in North Carolina Coastal Watersheds		16:00		IIID_Rulifson_03
Discrimination of <i>Sperata seenghala</i> stocks inhabiting three rivers of the Gangetic river system using elemental fingerprints on otoliths		16:05		IIID_Khan_04
Distinguishing Blacktip Shark, <i>Carcharhinus limbatus</i> , Nursery Areas in the Northern Gulf of Mexico with Vertebral Chemical Signatures		16:10		IIID_Lewis_06
Natal Origin of Pamlico Sound, North Carolina, Striped Bass Inferred from Otolith Chemistry		16:15		IIID_Dobbs_07
Using Otolith Shape Analysis as a tool of stock discrimination of the anchovy subspecies in the Black Sea		16:20		IIID_Sahin_08
Provenance and migration patterns of European whitefish <i>Coregonus lavaretus</i> (L.) s.l. in the Baltic Sea – combining otolith geochemistry and gill raker counts		16:25		IIID_Rohtla_09
Early life history of <i>Terapon jarbua</i> (Forsskål, 1775) using microstructures and Laser Ablation ICP-MS elemental composition of otoliths		16:30		IVD_Lavergne_08
Use of oxygen and carbon stable isotopes in otoliths to study fish movement and connectivity in <i>Lipophrys pholis</i>		16:35		IVD_Moreira_15
"Keeping earstones to the grindstones" POSTER SESSION AND WINE BAR		16:45 - 19:00	Poster Room	Theme and Workshop Posters on display all week

Wednesday, 22 October

AGE VALIDATION WORKSHOP			
Title	Time	Auditorium (Moderator) Presenter	Reference number
<i>Opening remarks</i>	09:00	(Richard McBride) Richard McBride	
Use of the stable oxygen isotope, 18O, in otoliths as an indicator of fish life history events and age validation	09:05	Craig Kestelle	WSAgeOral_ Kestelle_08
Refined bomb radiocarbon dating of two iconic fishes of the Great Barrier Reef	09:25	Allen Andrews	WSAgeOral_ Andrews_13
Advances in Ageing Techniques and Age Interpretation for U.S. West Coast Groundfish	09:55	Owen Hamel	WSAgeOral_ Hamel_10
<i>General discussion</i>	10:25		
Coffee Break	10:40		
Validation of age determination from Otoliths for the Anchovy in the Bay of Biscay.	11:00	(Ole Thomas Albert) Andres Uriarte	WSAgeOral_ Uriarte_06
Holistic approach on the age validation for the red mullet (<i>Mullus barbatus</i>) in the Southern Adriatic Sea (Central Mediterranean)	11:20	Pierluigi Carbonara	WSAgeOral_ Carbonara_15
A question of winter growth: Interpretation of Quarter 4 Celtic Sea whiting otoliths	11:40	Imelda Hehir	WSAgeOral_ Hehir_04
Can otolith weight be used as a trustworthy and quick predictor of age in <i>Pagellus erythrinus</i> (Pisces, Sparidae)?	12:00	Paraskevi Niki Lampri	WSAgeOral_ Lampri_09
Use and nonuse of tests of symmetry in age validation studies	12:20	Richard McBride	WSAgeOral_ McBride_03
<i>General discussion</i>	12:50		
Lunch	13:00		
Validation of annual zones in Greenland halibut otoliths from recaptures of chemically marked fish	14:30	(Allen Andrews) Ole Thomas Albert	WSAgeOral_ Albert_16
Age validation of monkfish (<i>Lophius americanus</i>)	14:50	Crista Bank	WSAgeOral_ Bank_11
Age and growth validation of the common thresher shark (<i>Alopias vulpinus</i>) in the northeastern Pacific Ocean	15:10	Natalie Spear	WSAgeOral_ Spear_02
Validation of the first annual increment deposition in the otoliths of European anchovy (<i>Engraulis encrasicolus</i> L.) in the Bay of Biscay	15:30	Naroa Aldanondo	WSAgeOral_ Aldanondo_05
Coffee Break	15:50		
Validation of daily increments in the otoliths of Atlanto-Iberian sardine larvae (<i>Sardina pilchardus</i> Walbaum, 1792) reared under different diets.	16:20	(Allen Andrews) Susana Ferreira	WSAgeOral_ Ferreira_07
Oral posters (10 min)	16:40 – 17:30	TBA	
<i>General discussion and concluding remarks</i>	17:30	Allen Andrews	

OTOLITH SHAPE ANALYSIS WORKSHOP		9:00 - 18:00	Seminar Room	
Title	Time	(Moderator) Presenter	Reference number	
Introduction to otolith shape analysis; theory behind the practice	09:00	(Audrey Geffen) Audrey Geffen		
Herring otolith shape – a tool for gauging stock complexity?	09:10	Lotte Worsøe Clausen	WSShape Worsøe Clausen_01	
Disentangling and quantifying sources of otolith shape variation across multiple scales using a new hierarchical partitioning approach	09:30	Matthias Vignon	WSShape_Vignon_04	
Morphological analysis applied to the shape of the sagitta of <i>Fundulus persimilis</i> (Cyprinodontidae).	09:40	Veronica Rivera	WSShape_Rivera_02	
Morphometric analysis of the sagitta otoliths of <i>Pterois volitans</i>	09:45	Jacob Rubio	WSShape_Rubio_03	
Otolith growth and shape changes during ontogeny in Atlantic bluefin tuna	09:50	Persefoni Megalofonou	WSShape_Megalofonou_06	
Discriminating stocks of common dentex (<i>Dentex dentex</i>) around Corsica Island (NW Mediterranean) using two otolith shape classification methods	09:55	Marie Baudouin	WSShape_Baudouin_07	
Image acquisition - code of best practice for obtaining high quality images with unambiguous outlines	09:55	Lotte Worsøe Clausen and Antoni Lombarte	WSShape Worsøe Clausen_08	
Coffee Break		10:40		
Image processing, outline generation and shape data extraction; interactive exercise	11:00	(Deirdre Brophy) Deirdre Brophy	WSShape_Brophy_09	
shapeR: an R package to study otolith shape variation	12:10	Lisa Anne Libungan	WSShape_Libungan_11	
What next? Introduction to analysing and interpreting otolith shape variation	12:30	Deirdre Brophy		
Lunch		13:00		
Assessment and avoidance of pitfalls by use of Fourier techniques in discrimination analysis of otolith contours from different stocks; interactive exercise	14:00	(Alf Harbitz) Alf Harbitz	WSShape_Harbitz_12	
Statistical analysis of population assignment for fisheries management employing otolith characteristics; interactive exercise	15:05	Henrik Mosegaard		
Coffee Break		15:45		

(CTD) Statistical analysis of population assignment for fisheries management employing otolith characteristics; interactive exercise	16:05	(Deirdre Brophy) Henrik Mosegaard	WSShape_ Mosegaard_13
Otolith identification system based on image contour analys	16:30	Youssef El Habouz	WSShape_ElHabouz__05
A conceptual modification of three Fourier techniques to represent 2D closed contours that requires only one frequency to reproduce a pure ellipse stocks	16:40	Alf Harbitz	WSShape_ Harbitz_14
Parameterization of 3D AFORO otolith surfaces	16:50	Pere Marti-Puig	WSShape Marti-Puig_10
Utility of Computer Assisted Age and Growth Estimation (CAAGE) in the analysis of otolith images for separating herring spawning groups	17:00	Mark Fisher	WSShape_ Fisher_15
Affordable 3D scanning of small otoliths for improved shape analysis by photogrammetry techniques	17:10	Andreas Zitek	WSShape_ Zitek_16
Morphometric analysis of the sagitta otoliths of Sciaenidae from Malaysia	17:20	Kar-Hoe Loh	WSShape_Loh_17
<i>Panel Discussion</i>	17:30		

Thursday, 23 October				
MORNING PLENARY SESSION	IIIA	9:00 - 13:10	Auditorium	Moderators C.Clemmesen, N.Aldanondo
Keynote Theme III: Population indicators		09:00	Auditorium	IIIA_Thorrold_Key (B.Morales-Nin speaker presentation)
Identifying the mechanisms shaping population structure in fish with dispersive life stages		9:45		IIIA_Wright_01
After two centuries of fish sclerochronology: the hegemony of otolith studies		10:05		IIIA_Panfili_02
Herring year classes after 1904 - how have they affected growth of subsequent year classes?		10:25		IIIA_Folkvord_03
Coffee grinder		10:45	Poster Room	
Untangling inter-stock differences in otolith d18O signatures: insights from a decade of plaice archival tagging.		11:15		IIIA_Darnaude_04
Stable isotopes in otoliths: what have we learned so far?		11:35		IIIA_Huijbers_05
Time and plaice: decadal changes in North Sea plaice <i>Pleuronectes platessa</i> L. observed through historical otolith and other legacy datasets		11:55		IIIA_Hunter_06
Operational viability of stock-separation using shape indices derived from the otolith morphometric outline. An example using sprat and herring.		12:15		IIIA_Mapp_07
Lunch luminance		12:35		
AFTERNOON PARALLEL SESSION	IIIB	14:00 - 15:40	Auditorium	Moderators P.Wright, K.Hüssy
Combined use of otolith shape, parasites and genetic markers for stock identification of the common dentex (<i>Dentex dentex</i>) around Corsica Island (NW Mediterranean).		14:00		IIIB_Marengo_01
The Baltic cod: A case study for testing stock discrimination based on otolith shape analysis in a mixed stock fishery		14:20		IIIB_Hüssy_02
Otolith shape variation in blue fin tuna from different regions of the North Atlantic: a possible marker of stock origin		14:40		IIIB_Brophy_03
Analysis of phenotypic characteristics of otoliths - resolving stock structure issues for snapper in South Australia.		15:00		IIIB_Fowler_04
Use of otolith quality flags to assess distributional dynamics in Baltic cod stock		15:20		IIIB_Stötera_05
AFTERNOON PARALLEL SESSION	IIIC	14:00 - 15:40	Seminar Room	Moderators C-H.Wang, P.Arechavala
Ecological changes in and recovery of the Ayu population following the tsunami generated by the 2011 Tohoku earthquake		14:00		IIIC_Kawakami_01
Out of sight, out of mind: what can otolith microstructure tell us about the elusive marine dispersive phase of the New Zealand whitebait <i>Galaxias maculatus</i> ?		14:20		IIIC_Egan_02

Pelagic larval duration and growth of early life stages of a coral reef fish: variations in an upwelling South Caribbean area		14:40		IIIC_Herrera-Reveles_03
Interpopulation and individual variation in dispersal characteristics of juvenile Sockeye Salmon		15:00		IIIC_Freshwater_04
Are larval reef fish travelling in packs? Using otoliths to evaluate evidence for shared dispersal histories.		15:20		IIIC_Shima_05
Coffee core		15:40	Poster Room	
AFTERNOON PARALLEL SESSION		16:10 - 17:30	Auditorium	Moderators E. Moksness , A.O. Thomas
Stable isotope otolith fingerprint signatures: a mass marking technique for farmed Atlantic salmon <i>Salmo salar</i>	IIIB	16:10		IIIB_Warren-Myers_06
Stable nitrogen isotopes in otoliths discriminate juvenile fish stocks in estuaries affected by anthropogenic impacts		16:30		IIIB_Lauchlan_07
Population Mixing between U.S. King Mackerel Stocks Estimated with Otolith Chemical Signatures		16:50		IIIB_Patterson_08
European flounder life history plasticity: paranormal activity or the helpful insights of otolith microchemistry?		17:10		IIIB_Morais_09
AFTERNOON PARALLEL SESSION		16:10 - 17:30	Seminar Room	Moderators M.Palmer, B.Morales
Dispersal capacities of Allis Shad (<i>Alosa alosa</i>) under global change: insights of innovative otolith microchemistry analysis	IIIC	16:10		IIIC_Martin_06
Investigating early stages dispersal using otolith chemistry: surprising relevance of the post-settlement phase in a temperate coastal fish.		16:30		IIIC_Di Franco_07
Assessing fish population connectivity: Combining otolith geochemistry and biophysical models		16:50		IIIC_Tanner_08
Estimating potential nursery sources for red snapper, <i>Lutjanus campechanus</i> , populations in Atlantic Ocean waters of the United States from North Carolina to Florida		17:10		IIIC_Barnett_09
OTOCHUKI		17:45 - 19:05	Auditorium	Moderator R.Vasconcelos
Are relationships between fish length and otolith radius enhanced by accounting for correlated errors? Implications for back-calculation.	IIID & IVD	17:45		IVD_Ashworth_01
Variability of the otolith growth rate and age of young-of-the-year chub, <i>Squalius cephalus</i> , from a hydraulic reach in the Rhône River.		17:50		IVD_Morat_02
Age and growth of John Dory <i>Zeus faber</i> (Linnaeus, 1758) in Atlantic Iberian waters using vertebrae and otolith increments		17:55		IVD_Piñeiro_04
Age, growth and distribution of the Antarctic fish <i>Pseudochaenichthys georgianus</i> NORMAN, 1939 in the Atlantic sector of Antarctic		18:00		IVD_Traczyk_04
Realizing connectivity- the influence of early life history on the dynamics of marine metapopulations		18:05		IIID_Swearer_05

Using the strontium isotope composition of otoliths from Bering cisco (<i>Coregonus laurettae</i>) to determine commercial stock composition in Alaska	18:15	IIID_Padilla_06
Morphometric analysis of yellowfin tuna (<i>Thunnus albacares</i>) otoliths in the western equatorial Atlantic Ocean	18:20	IIID_Batista da Silva_07
Competency phase affects patterns of fish early life traits	18:25	IVD_Calò_08
Residence depth of the deep-sea fish revealed by stable isotope file	18:30	IVD_Shiao_09
Use of otolith macrostructure for the identification of yearly birthday groups of European hake (<i>Merluccius merluccius</i>)	18:35	IIID_Carbonara_10
Outline variability of the saggita otoliths of the Patagonian grenadier (<i>Macruronus magellanicus</i>) in Chilean Patagonian waters	18:40	IIID_Cubillos_11
Fishery-dependent sampling may bias growth estimates	18:45	IIID_Morro_12
Life history Ba:Ca profiles reveal plasticity in the early life of the diadromous fish (<i>Centropomus undecimalis</i>)	18:50	IIID_Rolls_13
Daily age estimation of Japanese eel preleptocephali collected in the spawning area	18:55	IIID_Mochioka_15
Connectivity and habitat preference of the Brazilian weakfish (<i>Cynoscion acoupa</i>) by analysis of $\delta^{13}C$ of essential amino acids in otoliths	19:00	IIID_Vane_16

Friday, 24th October				
MORNING PLENARY SESSION	IVA	9:00 - 12:35	Auditorium	Moderators B.Morales-Nin, G.Petursdottir
Keynote Theme IV: Individual indicators		9:00	Auditorium	IVA_Grønkjær_Key (E.Moksness speaker presentation)
A multi-proxy approach for estimating estuarine immigration using otolith elements and tissue-specific stable isotopes		9:45		IVA_Mohan_01
Image-enhanced burns, bomb radiocarbon and microsatellite DNA improve the accuracy and precision of otolith-based age determinations for redfish (<i>Sebastes spp</i>)		10:05		IVA_Campana_02
Does the elemental composition of marine fish blood and otoliths reflect ambient conditions or physiology? Insights from the lab and the ocean		10:25		IVA_Sturrock_03
Coffee Antirostrum		10:45	Poster Room	
Maternal contribution process of Trace Elements in Striped Bass Otoliths During Early Ontogeny		11:15		IVA_Elking_04
Using otoliths to link population abundance decline to modifications in individual trophic niche and growth in a tropical fish species (<i>Bairdiella chrysoura</i> L., Sciaenidae)		11:35		IVA_Sirot_05
Less-than-daily growth increment formation in fish larvae otoliths: Exploring mechanisms with a bioenergetic modelling approach applied to Atlanto-Iberian sardine (<i>Sardina pilchardus</i>)		11:55		IVA_Pecquerie_06
Chemical signatures in scales reveal estuarine habitat use and trophic shifts of a highly migratory elopiform		12:15		IVA_Seeley_07
Lunch zone		12:35		
AFTERNOON PARALLEL SESSION		14:00 - 15:40	Auditorium	Moderators A.Fowler, A. Garcia
		14:00		
Dietary transmission of isotope spikes to otoliths, fin rays and scales: experimental validation and concentration-dependent mixing models		14:20		IVB_Walther_02
Sr isotope pattern deconvolution of LA-MC ICP-MS data to detect individual-specific transgenerational marks in freshwater fish otoliths		14:40		IVB_Irrgeher_03
Growth of stocked, marked eels compared to natural recruits in different habitats—a “common garden” approach		15:00		IVB_Wickström_04
Effects of temperature and ration on the otolith to body size relationship in juvenile Chinook salmon: A test of the direct proportionality assumption.		15:20		IVB_Stormer_05

AFTERNOON PARALLEL SESSION	IVC	14:00 - 15:40	Seminar Room	Moderators S.Thorrold, A.Darnaude
Natal signatures in the calcified structures of the giant Australian cuttlefish: comparing statoliths and cuttlebones		14:00		IVC_Woodcock_01
How long is the lifespan of European hake in the Mediterranean? Validating longevity		14:20		IVC_Vitale_02
Combining otolith microchemistry and microstructure analyses to infer transpacific migration patterns in juvenile Pacific bluefin tuna (<i>Thunnus orientalis</i>)		14:40		IVC_Baumann_03
Temperature history estimated with oxygen isotopes in otoliths of the Japanese common conger Conger myriaster collected from the continental shelf in the East China Sea.		15:00		IVC_Kawazu_04
Investigating diet and movement of cod (<i>Gadus morhua</i>) off Newfoundland using $\delta^{13}C$ of muscle and otolith amino acids		15:20		IVC_Piercey_05
Coffee Postrostrum		15:40	Poster Room	
AFTERNOON PARALLEL SESSION	IVB	15:40 - 17:20	Auditorium	Moderators L.Worsøe Clausen, D.Brophy
Assessment of otolith morphometrics as proxies for age in F-based assessments of tropical deepwater fisheries		16:10		IVB_Williams_06
Does diet influence otolith shape?		16:30		IVB_Mille_07
Otolith shape differences related to different migrating behavior in Icelandic cod tagged by Data Storage Tags		16:40		IVB_Bardarson_08
Otolith shape fluctuating asymmetry: A misconception of its biological relevance?		17:00		IVB_Díaz-Gil_09
AFTERNOON PARALLEL SESSION	IVC	15:40 - 17:20	Seminar Room	Moderators A.Folkvord, C.G.Piñeiro
Otolith examination in the seahorse <i>Hippocampus guttulatus</i>		16:10		IVC_Valladares_06
New open access software designed for the recognition of daily microstructures used in ageing sympatric hake species		16:30		IVC_Nava_07
Disentangling the effects of inherent otolith growth and model-simulated ecosystem parameters on the daily growth rate of young anchovies		16:40		IVC_Schismenou_08
Non-linear back-calculation in juvenile Baltic sprat (<i>Sprattus sprattus</i>): Insights into a recruitment-critical life stage		17:00		IVC_Günther_09
OTOCHUKI	IVD	17:30 - 18:45	Auditorium	Moderator P.Gaemers
A New Era of Ageing Spiny Dogfish (<i>Squalus suckleyi</i>) in the North Pacific Ocean		17:30		IVD_Tribuzio_01
Age structure of the goby <i>Parapocryptes serperaster</i> in the Mekong Delta, Vietnam, based on length-frequency and otolith analyses		17:35		IVD_Dinh_02
Changes in size, shape and appearance of juvenile Northeast Arctic cod (<i>Gadus morhua</i>) otoliths during settling		17:40		IVD_Irgens_03

Post-settlement growth of hake from the Norwegian coast based on otolith microstructure analysis: Evidence for counter gradient variation?	17:45	IVD_Staby_05
Colonization, hatch-dates, and growth rates of juvenile <i>Hyporhamphus picarti</i> (Actinopterygii, Hemiramphidae), in the Nador lagoon (NE Morocco)	17:50	IVD_Jaafour_06
Determination of strontium chemical environment and elemental mapping of otoliths by synchrotron radiation techniques	17:55	IVD_Dufour_07
Past and recent growth patterns in two species with contrasting demographic responses to environmental changes in a tropical lagoon	18:00	IVD_Sirot_08
The validation of otolith traceability tools within a robust, reproducible and transferable forensic framework	18:05	IIID_Geffen_10
Age and growth of larval Atlantic bluefin tuna, <i>Thunnus thynnus</i> , from the Gulf of Mexico	18:10	IVD_Malca_09
Shape analysis of sagittal otolith of <i>Haemulon plumierii</i> caught in Pernambuco State (Brazil): ontogenetic and population perspectives	18:15	IVD_Vasconcelos Filho_10
Larval Otolith Formation in Striped Bass (<i>Morone saxatilis</i>)	18:20	IVD_Elking_11
Using otolith strontium isotopes to reconstruct life history portfolios within salmon populations: When do different phenotypes contribute?	18:25	IVD_Sturrock_12
Migration patterns and population structure of two small pelagic species from otolith microchemistry: European anchovy (<i>Engraulis encrasicolus</i>) and Sardine (<i>Sardina pilchardus</i>) of the Bay of Biscay	18:30	IIID_Gatti_13
Contribution of otolith microchemistry to reveal the evolutionary origin of catadromous migration in anguilliformes fishes: an example of tropical moray eel	18:35	IVD_Kuroki_14
Age estimation and otolith analysis of <i>Fistularia commersonii</i> Rüppell, 1838 (Syngnathiformes – <i>Fistularia</i>) in the central Mediterranean Sea	18:40	IVD_Vitale_16
Otolith microchemistry: A useful tool for age validation?	18:45	IVD_Hüssy_17
Can we derive a deep water $\delta^{14}\text{C}$ curve to aid age validation of NZ deep sea fish species?	18:50	IVD_Neil_18
SYMPOSIUM CLOSING SESSION	19:15	Auditorium
Demons - "A little asymmetry goes a long way"	19:30	Casal Grounds
GALA DINNER: Hard Rock Story's happily ever-afters	20:00	Hotel Palmira Beach

Theme I: Environmental Indicators

Poster Session (IE)

Abstract reference: IE_Labonne_01

Characterization of otolith microchemical signatures from 3 fish species along the Moroccan coast: link between anthropogenic vs. natural influences and trophic level

Labonne, M.; Masski, H.; Tai, I.; Lae, R.; Bouthir, F.Z.; Bassoullet, C.; Tito de Morais, L.

Contact e-mail: maylis.labonne@ird.fr

Abstract reference: IE_Janowska_02

Chemical composition of fish otoliths from a lake subject to reclamation

Heese, T.; Lampart-Kałużniacka, M.; Janowska, B.; Siebielska, I.

Contact e-mail: beata.janowska@tu.koszalin.pl

Abstract reference: IE_Reis-Santos_03

Effects of temperature and water composition on otolith chemistry across a salinity gradient

Reis-Santos, P.; Tanner, S.E.; Cabral, H.N.; Gillanders, B.M.

Contact e-mail: pnsantos@fc.ul.pt

Abstract reference: IE_Limburg_04

Fish Tales Through Fish Ears

Limburg, K.

Contact e-mail: klimburg@esf.edu

Abstract reference: IE_Agiadi_05

How did past environmental change control the distribution of small pelagic fish in the Mediterranean Sea? Examples from the fossil record

Agiadi, K.; Karakitsios, V.

Contact e-mail: kagiadi@geol.uoa.gr

Abstract reference: IE_Songer_06

Investigating the distribution of crystalline otoliths

Songer, S.; Smith, M.

Contact e-mail: sally.songer@cefas.co.uk

Abstract reference: IE_Górski_07

Migratory life-history patterns of *Galaxias maculatus* in the Southern Hemisphere rivers revealed by otolith microchemistry

Górski, K.; Habit, E.M.; Manosalva, A.J.

Contact e-mail: kgorski@udec.cl

Abstract reference: IE_Helser_08

Modelling Environmental Factors Affecting Assimilation of Bomb-produced $\Delta^{14}\text{C}$ in the North Pacific Ocean: Implications for age validation studies

Helser, T.E., Kastle, C.R.; Lai, H.

Contact e-mail: thomas.helser@noaa.gov

Abstract reference: IE_Paulus_09

Namibian hake *M. capensis* otolith zonation related to fish and environmental conditions in the northern Benguela

Paulus, S.C.; Wilhelm, M.R.; Kashava, S.; Shivute, L.

Contact e-mail: sharlie916@hotmail.com

Abstract reference: IE_McFadden_10

Otolith Biomineralisation: Insights From a Microstructural and Microanalytical Study

McFadden, A.; Gillanders, B.M.; Pring, A.; Wade, B.

Contact e-mail: bronwyn.gillanders@adelaide.edu.au

Abstract reference: IE_Arechavala-Lopez_11

Otoliths as indicators of wild or farm origin of sea bream and sea bass

Arechavala-Lopez, P.; Pablo Sanchez-Jerez, P.

Contact e-mail: pablo.arechavala@ua.es

Abstract reference: IE_Limburg_12

Paleo- and modern salinity and temperatures as recorded by Baltic Sea cod *Gadus morhua* over millennia

Limburg, K.E.; Olson, C.

Contact e-mail: klimburg@esf.edu

Abstract reference: IE_Fabré_13

Pattern of otolith growth in earlier stages of *Mugil liza* in tropical and temperate regions of Southwestern Atlantic Ocean

Fabré, N. N.; Ferreira, S.; Fortunato, M.; Batista, C.R.; Batista, V.S.; Volpedo, A.V.

Contact e-mail: tchoni1@uol.com.br, avolpedo@fvet.uba.ar

Abstract reference: IE_Evans_14

Tracking Stocked European Eel (*Anguilla anguilla*) using Otolith Microchemistry

Evans, D.; Bartkevics, V.; Wickstrom, H.

Contact e-mail: derek.evans@afbini.gov.uk

Abstract reference: IE_Volpedo_15

Use of lapillus otolith microchemistry as indicator of the habitat of *Genidens barbuis* in different estuarine environments from the southwestern Atlantic Ocean

Avigliano, E.; Velasco, G.; Volpedo, A.V.

Contact e-mail: avolpedo@gmail.com

Abstract reference: IE_Heimbrand_16

Using otoliths to distinguish sea spawning from river spawning whitefish (*Coregonus mareana*)

Heimbrand, Y.; Odelström, A.; Elfman, M.; Florin, A.B.

Contact e-mail: yvette.heimbrand@slu.se

Abstract reference: IE_Buckthought_17

Utility of otolith chemistry in determining stock structure of grey mullet (*Mugil cephalus*) in northern New Zealand

Buckthought, D.

Contact e-mail: dane.buckthought@niwa.co.nz

Abstract reference: IE_Izzo_18

Where do elements bind within the otoliths of fish?

Izzo, C.; Doubleday, Z.A.; Gillanders, B.M.

Contact e-mail: c.izzo@adelaide.edu.au

Slideshow Poster Session (ISS)

Abstract reference: ISS_Tracey_01

Age and growth study of deepsea coral in aquaria

Cummings, V.; Neil, H.; Barr, N.; Moss, G.; Marriott, P.; Davy, S.; Gammon, M.

Contact e-mail: di.tracey@niwa.co.nz

Abstract reference: ISS_Plaza_02

Chronological growth patterns of the endolithic bivalve *Petricola patagonica* associated to a subduction Chilean earthquake

Plaza, G.; Cisternas, M.; Melnick, D.

Contact e-mail: guido.plaza@ucv.cl

Abstract reference: ISS_Hällbom_03

The importance of age determination for assessment of the status in fish fauna

Ardestam, B.; Hällbom, M.; Holmgren, K.

Contact e-mail: malin.hallbom@slu.se

Abstract reference: ISS_Walker_04

The use of otoliths to compare three species from the Zoarcid family

Walker, K.; Norcross, B.

Contact e-mail: klwalker2@alaska.edu

Abstract reference: ISS_Wang_05

Tracing the impact of oil spills event by using chemical signatures in squid statoliths and tissues

Wang, T.; Hsu, Z.; Wang, C.

Contact e-mail: sw14916@hotmail.com

Theme II: Community Indicators

Poster Session (IIE)

Abstract reference: IIE_QUIJANO-Puerto_01

Age and growth of the invasive lionfish (*Pterois volitans*) in the Parque Nacional Arrecife Alacranes, Southern Gulf of Mexico

Quijano-Puerto, L.; Aguilar-Perera, A.

Contact e-mail: luis.quijano18@gmail.com

Abstract reference: IIE_Mahouachi_02

Application of otolith shape analysis in identifying different species of *Zosterisessor ophiocephalus* and *Gobius paganellus* in the lagoon of Bizerte in Tunisia

Mahouachi, N.H.; Maazi, F.; Marsaoui, B.; Fatnassi, M.; Trojette, M.; Khedher, M.; Messaoud, R.; Rebaya, M.; Chalah, A.; Quignard, J.P.; Trabelsi, M.

Contact e-mail: marsaouibochra@gmail.com

Abstract reference: IIE_Curin-Osorio_03

Comparative morphometry of the sagitta otolith in *Sardina Común* (*Strangomera bentincki*) and *Sardina austral* (*Sprattus fuegensis*) in southern Chile

Curin-Osorio, S.

Contact e-mail: sacurin@gmail.com

Abstract reference: IIE_Brind'Amour_12

Combining biological markers to estimate the realized niche species: the case of three flatfish in the Seine estuary

Brind'Amour, A.

Contact e-mail: durieux@univ-corse.fr

Abstract reference: IIE_Sousa_13

Ecomorphological patterns of the sagittae otolith in tropical fish of the western south Atlantic

de Sousa, M.F.; Fabré, N.N.; Volpedo, A.D.; da Silva, J.P.

Contact e-mail: marcia_ufal@hotmail.com

Abstract reference: IIE_Tuset_13

Sagittal otolith morphology helps to explain the invasion success of Lessepsian species

Víctor M. Tuset¹, **Antoni Lombarte**¹, Michel Bariche², Francesc Maynou¹, Ernesto Azzurro³

¹Institut de Ciències del Mar (CSIC). Barcelona, Catalonia, SPAIN

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³ISPRA, National Institute for Environmental Protection and Research, ITALY

Contact e-mail: vtuset@icm.csic.es

Abstract reference: IIE_Lampart-Kaluzniacka_14

Effect of disturbances in the structure of trophic lakes on the formation of annual marks in predatory and planktivorous fish

Lampart-Kałużniacka, M.; **Heese, T.**

Contact e-mail: mlampart@tu.koszalin.pl

Abstract reference: IIE_Callicó Fortunato_15

Identification of Mediterranean Mulletts using morphology of sagittae otoliths

Callicó Fortunato, R.; Durà,V.B.; Volpedo., A.

Contact e-mail: roberta_cali@yahoo.com.ar

Abstract reference: IIE_Durieux_17

Insights on otolith shape diversity of Mediterranean postlarval fishes

Durieux, E.

Contact e-mail: durieux@univ-corse.fr

Abstract reference: IIE_Gerard_18

Isotopic signatures in the otoliths of reef-associated fishes of southern Florida: linkages between nursery grounds and coral reefs

Gerard, T.; Malca, E.; Muhling, B.; Mateo, I.; Lamkin, J.

Contact e-mail: trika.gerard@noaa.gov

Abstract reference: IIE_Shen_19

Migration history comparison among three *Mugil cephalus* cryptic species in Northwest Pacific revealed from otolith Sr/Ca ratio

Shen, K.

Contact e-mail: knshen@mail.ntou.edu.tw

Abstract reference: IIE_Gholami_20

Morphology of the sacular otoliths in species identification, phylogeny and systematic of the Iranian *Aphanius* (Cyprinodontidae; Cyprinodontidae) species

Gholami, Z.; Reichenbacher, B.; Esmaeili, H.R.; Teimori, A.

Contact e-mail: zgholami2005@gmail.com

Abstract reference: IIE_Godiksen_21

Otoliths as life history indicators

Godiksen, J.

Contact e-mail: jane.godiksen@imr.no

Abstract reference: IIE_Wang_22

Spatial-temporal distribution and migratory pattern among mullet species in Tanshui River

Wang, C.

Contact e-mail: chwang99@mail.ntou.edu.tw

Slideshow Poster Session (IISS)

Abstract reference: IISS_Chang_01

Using otolith morphology in zoogeography and phylogeny analyses of Sciaenidae

Chang, M.Y.; Chao, N.L.; Shao, K.T.; **Chang, C.W.**

Contact e-mail: mychang@most.gov.tw

Abstract reference: IISS_Chang_02

Fish otolith assemblages from Tainan Science Park archaeological sites in southwestern Taiwan

Lin, C.H.; Li, K.T.; **Chang, C.W.**

Contact e-mail: changcw@nmmba.gov.tw

Abstract reference: IISS_Lombarte_03

Identifying Mediterranean Sea gobies using otoliths

Lombarte, A.; Miletić, M.; Kovačić, M.; Otero-Ferrer, J.L.; Tuset, V.M.

Contact e-mail: toni@icm.cat

Abstract reference: IISS_Songer_04

Maintaining quality standards throughout the collection, processing, reading and archiving of otoliths

Songer, S.

Contact e-mail: sally.songer@cefias.co.uk

Abstract reference: IISS_Khan_05

Otolith research in India: status and perspective

Khan, M.A.; Kaish Miyan

Contact e-mail: khanmafzal@yahoo.com

Abstract reference: IISS_Mahouachi_06

Otolithometry aspect of differentiation of *Trachurus trachurus* (Perciformes, Carangidae) in the lagoon of Bizerte and the island of Galite in Tunisia

Mahouachi, N.; Maazi, F.; Marsaoui, B.; Fatnassi, M.; Trojette, M.; Kheder, M.; Messaoud, R.; Rebaya, M.; Ben Alaya, H.; Chalah, A.; Quignard, J.; Trabelsi, M.

Contact e-mail: marsaouibochra@gmail.com

Abstract reference: IISS_Rossi-Wongtschowski_07

Teleostei Fish Otoliths from the S-SE Brazil: a sagittae collection, its website and publications

Rossi-Wongtschowski, C.L.D.B.; Siliprandi, C.C.; Chalom, A.; Brenha, M.R.; Vaz-dos-Santos, A.M.

Contact e-mail: cwongski@usp.br

Abstract reference: IISS_Reichenbacher_08

The Late Miocene freshwater fish fauna from Venta del Moro (Valencia, Spain)

Reichenbacher, B.; Montoya, P.

Contact e-mail: b.reichenbacher@lrz.uni-muenchen.de

Theme III: Population Indicators

Poster Session (IIIE)

Abstract reference: IIIE_Sheridan_01

A potential method for the age determination of Ireland's Norway lobster (*Nephrops norvegicus*) populations using calcified structures.

Sheridan, M.; Officer, R.; O'Connor, I.; Lordan, C.

Contact e-mail: michael.sheridan@research.gmit.ie

Abstract reference: IIIE_Condini_02

Age and growth of Dusky Grouper (*Epinephelus marginatus*) (Perciformes: Epinephelidae) from its southernmost population in Southwestern Atlantic, with a size comparison between offshore and littoral habitats

Condini, M.

Contact e-mail: mvcondini@gmail.com

Abstract reference: IIIE_Rezende_03

Age and Growth of Snapper Species (Lutjanidae) from the Tropical Southwestern Atlantic Ferreira, B.P.; Rezende, S.M.

Contact e-mail: rezende_sergiomagalhaes@hotmail.com

Abstract reference: IIIE_Egan_04

Can otolith shape discriminate between regional populations of a widely dispersing New Zealand diadromous fish?

Egan, E.; Hickford, M.; Quinn, J.; Schiel, D.

Contact e-mail: eimear.egan@pg.canterbury.ac.nz

Abstract reference: IIIE_Farias_05

What new insights can otolith microchemistry and shape analysis bring to the black scabbardfish, *Aphanopus carbo*, migratory hypothesis?

Farias, I.; Pérez-Mayol, S.; Palmer, M.; Figueiredo, I.; Morales-Nin, B.

Contact e-mail: ifarias@ipma.pt

Abstract reference: IIIE_Fatnassi_06

Comparing otolith shape for stock discrimination of *Trachinus draco* (Trachinidae) in Tunisian waters

Fatnassi, M.; Mahouachi, N.H.; Trojette, M.; Marsaoui, B.; Ben Alaya, H.; Chalah, A.; Quignard, J.P.; Trabelsi, M.

Contact e-mail: manel.ft@gmail.com

Abstract reference: IIIE_Messaoud_07

Otolithometric comparative study of two Tunisian lagoons population of *Liza aurata* (Lagoon Ghar El Melh and Ichkeul)

Messaoud, R.; Marsaoui, B.; Fatnassi, M.; Trojette, M.; Chalah, A.; Khedher, M.; Rebaya, M.; Ben Alaya, H.; Quignard, J.; Trabelsi, M.

Contact e-mail: marsaouibochra@gmail.com

Abstract reference: IIIE_Khedher_08

Importance of otolith morphology in the discrimination of two Tunisian populations of *Mugil cephalus* (Sea of Tabarka and Ghar El Melh Lagoon)

Kheder, M.; Fatnassi, M.; Trojette, M.; Marsaoui, B.; Rebaya, M.; Messaoud, R.; Mahouachi, N.; Chalah, A.; Quignard, J.; Trabelsi, M.

Contact e-mail: khedher.maissa@gmail.com

Abstract reference: IIIE_Marsaoui_09

Use of the contour shape of otoliths in the discrimination of two batches of sea bream (*Sparus aurata*) wild and farmed fish

Marsaoui, B.; Kheder, M.; Messaoud, R.; Fatnassi, M.; Trojette, M.; Rebaya, M.; Ben Alaya, H.; Chalah, A.; Quignard, J.; Trabelsi, M.

Contact e-mail: marsaouibochra@gmail.com

Abstract reference: IIIE_Saygili_09

Comparison of otolith morphometric characteristics of *Spicara maena* in the north Aegean sea and sea of Marmara

Saygili, B.; Ismen, A.; Arslan, M.

Contact e-mail: buraksaygili@gmail.com

Abstract reference: IIIE_Diouf_10

Connectivity between populations of the threatened white grouper (*Epinephelus aeneus*) along the West-African coasts inferred by coupling otolith multi-elemental composition and genetic analyses

Agiadi, K.; Karakitsios, V.

Contact e-mail: khady1.diouf@ucad.edu.sn

Abstract reference: IIIE_Kokkin_11

Consequences of within-lake differences in life-history characteristics of pikeperch

Kokkin, M.

Contact e-mail: magnus.kokkin@slu.se

Abstract reference: IIIE_Arula_12

Discrimination of herring ecotypes using otolith shape and body morphology in the Baltic Sea

Arula, T.; Shpilev H.; Raid, T.

Contact e-mail: timo.arula@ut.ee

Abstract reference: IIIE_Svirgsden_13

Do Eurasian minnows (*Phoxinus phoxinus*) inhabiting brackish water enter freshwater to reproduce: evidence from a study on otolith microchemistry

Svirgsden, R.; Rohtla, M.; Albert, A.; Taal, I.; Vetemaa, M.

Contact e-mail: roland.svirgsden@ut.ee

Abstract reference: IIIE_Condini_14

Dusky groupers from southern Brazil: can littoral fish groups be supplied by neritic populations?

Condini, M.; Queiroz de Albuquerque, C.; Miranda Garcia, A.

Contact e-mail: mvcondini@gmail.com

Abstract reference: IIIE_Martinho_15

Early life stages of plaice *Pleuronectes platessa* in cold-water nurseries

Martinho, F.; Freitas, V.; Santos, P.; Bremm, C.; Campos, J.; van der Veer, H.W.

Contact e-mail: fmdm@ci.uc.pt

Abstract reference: IIIE_Dwyer_16

Effect of ageing errors on population dynamics from analytical assessments for two species of flatfish in the Northwest Atlantic

Dwyer, K.S.; Morgan, M.J.; Healey, B.P.

Contact e-mail: Karen.Dwyer@dfo-mpo.gc.ca

Abstract reference: IIIE_Rebaya_16

Importance of sagittal otolith shape in discrimination stocks *Liza ramada* two locations (Sea Cap zebib and Dam Mellegue) Tunisia

Rebaya, M.; Fatnassi, M.; Marsaoui, B.; Khedher, M.; Messaoud, R.; Mahouachi, N-H.; Ben Alaya, H.; Chalah, A.; Quignard, J-P.; Trabelsi, M.

Contact e-mail: marsaouiibochra@gmail.com

Abstract reference: IIIE_Dou_17

Effects of elemental concentration and fish growth on strontium and barium incorporation into otoliths of larval and juvenile flounder *Paralichthys olivaceus*

Dou, S.

Contact e-mail: szdou@qdio.ac.cn

Abstract reference: IIIE_Jurado-Ruzafa_19

First approach to the growth of *Trachurus picturatus* (Bowdich, 1825) from the Canary Islands (Spain)

Jurado-Ruzafa, A.; Santamaría, M.T.G.

Contact e-mail: alba.jurado@ca.ieo.es

Abstract reference: IIIE_Ariza_20

Growth And Age Of The Indo-Pacific Lionfish *Pterois Volitans* (Pisces: Scorpaenidae), In Waters Of Venezuelan Central Coast, South Caribbean

Ariza, L.A.; Núñez, J.G.; Herrera-Reveles, A.T.; Narváez, M.; Martínez, A.T.; Marín, B.

Contact e-mail: luisalejandroariza@gmail.com

Abstract reference: IIIE_Lozys_21

Habitat use and migratory behavior of pikeperch *Sander lucioperca* in Lithuanian and Latvian waters as inferred from otolith Sr:Ca ratios

Ložys, L.; Shiao, J.C.; Iizuka Y.; Minde, A.

Contact e-mail: jcshiao@ntu.edu.tw

Abstract reference: IIIE_Cerna_22

High growth of the Peruvian anchovy (*Engraulis ringens*) in northern Chile, estimated using daily increment of sagittal otoliths in juvenile and adult fish

Cerna, F.; Plaza, G.

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Abstract reference: IIIE_Shivute_23

Horse mackerel *Trachurus capensis* age validation in the northern Benguela

Shivute, L.

Contact e-mail: lshivute@mfmr.gov.na

Abstract reference: IIIE_Duncan_24

Investigation of the population and sub-population structure of Albacore tuna in the Northeast Atlantic and Mediterranean

Duncan, R.; Brophy, D.; Arrizabalaga, H.; Tinto, F.

Contact e-mail: roxanne.duncan@gmit.ie

Abstract reference: IIIE_Turner_25

Juvenile river herring habitat use and emigration trends throughout their U.S. ranges

Turner, S.; Limburg, K.

Contact e-mail: smturner483@gmail.com

Abstract reference: IIIE_Yang_26

Life history of three conger eel species in the waters of Northeast Taiwan

Yang, Y.; Wang, W.

Contact e-mail: yihua802@yahoo.com.tw

Abstract reference: IIIE_Santos-Cruz_28

Morphometric description of sagittae otoliths to differentiate two morphotypes of white mullet *Mugil curema* (Mugilidae) in the Santos bay-estuary complex (Brazil)

Santos-Cruz, N.N.; Souza, M.R.; Tomás, A.R.G.

Contact e-mail: nayranicolaubio@yahoo.com.br

Abstract reference: IIIE_Ferraton_29

Optimising LA-ICPMS rastering protocols and data reduction procedures to produce otolith micro-chemical signatures allowing robust reconstruction of fish past habitats

Ferraton, F.; Sirot, C.; Guillaumon, F.; Tournois, J.; Childs, A.; Darnaude, A.

Contact e-mail: franck.ferraton@univ-montp2.fr

Abstract reference: IIIE_Karakulak_30

Otolith Characteristics of Annular Sea Bream (*Diplodus annularis*) in northern Aegean Sea (Turkey)

Yıldız, T.; Uzer, U.; Karakulak, F.S

Contact e-mail: karakul@istanbul.edu.tr

Abstract reference: IIIE_Kitchens_31

Otolith chemical signatures discriminate between nursery areas of yellowfin tuna (*Thunnus albacares*) in the Atlantic Ocean

Kitchens, L.; Rooker, J.

Contact e-mail: l.kitchens@tamu.edu

Abstract reference: IIIE_Zanella_32

Otolith morphology and growth of a short-lived freshwater goby

Zanella, D.; Miletić, M.; Mrakovčić, M.; Bermanec, V.; Mustafić, P.; Čaleta, M.; Marčić, Z.; Buj, I.; Mihinjač, T.

Contact e-mail: davor.zanella@zg.t-com.hr

Abstract reference: IIIE_Maciel_33

Otolith relative growth: a tool to identify sexual dimorphism in the Guri Sea Catfish *Genidens genidens*

Maciel, T.R.; Vaz-dos-Santos, A.M.; Vianna, M.

Contact e-mail: andrevaz@ufpr.br

Abstract reference: IIIE_Nachón_34

Otolith's elemental and isotopic profiles of twaite shad [*Alosa fallax* (Lacépède, 1803)]: insight into habitat use and discriminate stocks.

Nachón, D.J.; Mota, M.; Antunes, C.; Bareille, G.; Pecheyran, C.; Vieira-Lanero, R.; Cobo, F.

Contact e-mail: davidjose.nachon@usc.es

Abstract reference: IIIE_Araya_35

Partial migration in introduced wild chinook salmon (*Oncorhynchus tshawytscha*) of southern Chile

Araya, M.; Niklitschek, E.J.; Secor, D.H.; Piccoli, P.M.

Contact e-mail: maraya@unap.cl; miguelaray@gmail.com

Abstract reference: IIIE_Wells_36

Population structure of albacore (*Thunnus alalunga*) in the eastern Pacific Ocean

Wells, R.J.D.; Kohin, S.; Dewar, H.; Kinney, M.J.; Rooker, J.R.; Snodgrass, O.R.

Contact e-mail: wells@tamug.edu

Abstract reference: IIIE_Paillon_37

Quantifying the role of mangroves for the blackspot snapper (*Lutjanus fulvivflamma*) by otolith microchemistry and UVC, in a South-Pacific archipelago (New-Caledonia)

Paillon, C.

Contact e-mail: christelle.paillon@ird.fr

Abstract reference: IIIE_Tabouret_38

Reading the environment before reading the otolith: a key to tell *Sicyopterus lagocephalus* stories in La Réunion Island rivers

Tabouret, H.; Teichert, N.; Lagarde, R.; Holub, A.; Barbotin, G.; Grondin, H.; Pécheyran, C.; Bareille, G.

Contact e-mail: helene.tabouret@univ-pau.fr

Abstract reference: IIIE_Libungan_39

shapeR: an R package to study otolith shape variation

Libungan, L.

Contact e-mail: lal@hi.is

Abstract reference: IIIE_Nash_40

Spatial linkages in the early life history of north eastern Atlantic herring populations across the north of the British Isles.

Nash, R.D.M.; Payne, M.R.; Geffen, A.J.

Contact e-mail: richard.nash@imr.no

Abstract reference: IIIE_Trojette_41

Study of the morphological variation of the otolith of fish (*Scorpaena Porcus*) between a marine environment (Hammam-Lif) and a middle island (Djerba) in Tunisia

Trojette , M.; Fatnassi, M.; Marsaoui, B.; Khedher, M.; Rebaya, M.; Messaoud, R.; Mahouachi, N.; Ben Alaya, H.; Chalah, A.; Quignard, J.; Trabelsi, M.

Contact e-mail: marsaouibochra@gmail.com

Abstract reference: IIIE_Frouzova_42

The growth of perch (*Perca fluviatilis*) in newly filled post mining lake

Frouzova J.; Tumova E.; Vejrik L.; Peterka J.

Contact e-mail: jarkafrouzova@gmail.com

Abstract reference: IIIE_Santana_43

The importance of the otoliths microstructures in the adjustment of parameters and description of growth in fishes

Lessa, R.; **Santana, F.M.**

Contact e-mail: framarsan@ig.com.br

Abstract reference: IIIE_Khemiri_44

The use of otolith shape to determine stock structure of *Sardina pilchardus* and *Engraulis encrasicolus* in Tunisian coasts

Khemiri, S.

Contact e-mail: sanak182000@yahoo.com

Abstract reference: IIIE_Mahé_45

The use of otolith shape to evaluate the stock structure of swordfish (*Xiphias gladius*) in the Indian Ocean

Mahé, K.; Evano, H.; Mille, T.; Bourjea, J.

Contact e-mail: kelig.mahe@ifremer.fr

Abstract reference: IIIE_Westgaard_46

The use of otoliths as DNA source in population genetics: population structure of European hake by the use of SNPs

Westgaard, J-I; **Godiksen, J.A.**; Staby, A.; Svedäng, H.; André, C; Geffen, A.J.

Contact e-mail: jon-ivar.westgaard@imr.no

Abstract reference: IIIE_Kemp_47

Trace element mapping of Bight redfish otoliths to assess stock structure in Western Australia

Kemp, J.; Jackson, G.; Norriss, J.; Evans, N.; Taylor, R.; McDonald.

Contact e-mail: jodie.kemp@fish.wa.gov.au

Abstract reference: IIIE_Ruas_48

Updating the age and growth of the rough scad, *Trachurus lathami* Nichols, 1920 in the Southwestern Atlantic

Ruas, L.; **Vaz-dos-Santos, A.M.**

Contact e-mail: andrevaz@ufpr.br

Abstract reference: IIIE_Moreira_49

Use of otolith shape analysis for population structure study of the coral reef fish *Stegastes fuscus* (Pomacentridae) from coastal islands of south Brazil

Daros, F.; Tuset, V.M.; Otero-Ferrer, J.L.; **Moreira, C.**; Spach, H.; Correia, A.T.

Contact e-mail: claudia.moreira@ciimar.up.pt

Abstract reference: IIIE_Rutterford_51

Using otoliths to gauge sub-population responses to warming seas

Rutterford, L.A.; Hunter, E.

Contact e-mail: lar210@exeter.ac.uk

Abstract reference: IIIE_Geffen_52

Vaster than empires....but more fast: large-scale otolith analyses in population and traceability studies (the FishPopTrace collection)

Geffen, A.J.; The FishPopTrace Consortium

Contact e-mail: audrey.geffen@bio.uib.no

Abstract reference: IIIE_Hägerstrand_53

Whitefish (*Coregonus lavaretus*) groups in the brackish northern Baltic Sea discriminated by otolith elemental bulk analysis

Hägerstrand, H.; Himberg, M.; Numers, M.; Mrowczynska, L.; Jokikokko, E.; Vasemägi, A.; Wiklund, T.; Lill, J-O.

Contact e-mail: hhagerst@abo.fi

Slideshow Poster Session (IISS)

Abstract reference: IISS_Vasconcelos Filho_01

Ecomorphology of sagitta from *Plagioscion squamosissimus* (Heckel, 1840)

Vasconcelos-Filho, J.E.; Costa, R.S.; Lessa, R.P.T.

Contact e-mail: jonas.vasconcelos.filho@gmail.com

Abstract reference: IISS_Pilinlovskij_02

Fish-passes in existing dams of Kražantė and Sausdravas rivers and pass reconstruction in river Vilnia, radio telemetry and conventional tagging studies for Atlantic sturgeon *Acipenser oxyrinchus* Mitchill post release migration

Pilinkovskij, A.

Contact e-mail: Justas.poviliunas@zuv.lt

Abstract reference: IISS_Zuykova_03

Impact of bias in age reading of the Northeast Arctic cod on stock assessment

Zuykova, N.; Yaragina, N.; Kovalev, Y.; Chetyrkin, A.

Contact e-mail: zunat@pinro.ru

Abstract reference: IISS_Silva_04

Morphology of the sagittal otolith of the Whitemouth croaker, *Micropogonias furnieri*, and of the Smooth weakfish, *Cynoscion leiarchus*, in the Southwestern Atlantic, Brazil

Silva, J.P.C.; Rodrigues da Costa, M.; Vaz-dos-Santos, A.M.; Santos, R.S; Oliveira, P.N.

Contact e-mail: jose_paulo_cs@hotmail.com

Abstract reference: IIISS_Bal_05

Otolith Biometry - Total Length Relationships in the Population of Hazar Bleak, *Alburnus heckeli* (Battalgi, 1943) Inhabiting Lake Hazar, Elazig, Turkey

Bal, H.

Contact e-mail: hatice.bal@hotmail.com

Abstract reference: IIISS_Ching_06

Spatial and temporal variation in statolith elemental signatures of the *Sepioteuthis lessoniana* around northern Taiwan

Ching, T.; Chen, C.; Wang, C.

Contact e-mail: star790402@hotmail.com

Abstract reference: IIISS_Walsh_07

The development of fish age determination protocols for commercially important New Zealand species

Walsh, C.

Contact e-mail: cameron.sms@xtra.co.nz

Abstract reference: IIISS_Farias_08

Using otoliths to identify *Aphanopus carbo* and *Aphanopus intermedius*

Farias, I.; Pérez-Mayol, S.; Palmer, M.; Morales-Nin, B.; Figueiredo, I.

Contact email: ifarias@ipma.pt

Theme IV: Individual Indicators

Poster Session (IVE)

Abstract reference: IVE_Sainza_01

Age and Growth of European conger eel (*Conger conger*) in the North East Atlantic Ocean using otoliths and vertebrae.

Sainza, M.

Contact e-mail: maria.sainza@vi.ieo.es

Abstract reference: IVE_Matić-Skoko_03

Age determination and validation on otoliths of the striped red mullet, *Mullus surmuletus* from the Adriatic Sea

Matić-Skoko, S.; Ferri, J.; Brajčić Jurica, D.

Contact e-mail: sanja@izor.hr

Abstract reference: IVE_Vittori_04

Age estimation and back-calculation of fish length of the European hake, *Merluccius merluccius* (Linnaeus, 1758) in Sardinian waters.

Vittori, S.; Bellodi, A.; Agus, B.; Consolo, A.; Soldovilla, G.; Follesa, M.C.; Pesci, P.

Contact e-mail: svittori@unica.it

Abstract reference: IVE_Jean-Christophe_05

Age estimation of the brown trout, *Salmo trutta* L introduced in the Kerguelen Islands: Determination of the otolith optimal zone for annuli detection.

Aymes, J.C.; Vignon, M.; Beall, E.; Guéraud, F.; Gaudin, P.

Contact e-mail: jcaymes@st-pee.inra.fr

Abstract reference: IVE_Ventero_06

Anchovy (*Engraulis encrasicolus*) otoliths reveal growth differences between two areas of the Spanish Mediterranean Sea

Ventero, A. , Iglesias, M. , Villamor, B.

Contact e-mail: aventero@ba.ieo.es

Abstract reference: IVE_Ferri_07

Assessing the use of otolith morphometrics to predict the age of the black scorpionfish, *Scorpaena porcus* (Linnaeus, 1758)

Ferri, J.; Brčić, J.; Matić-Skoko, S; Škeljo, F.

Contact e-mail: josipa.ferri@unist.hr

Abstract reference: IVE_Carvalho_10

Using otolith microchemistry as a tool on migration studies of *Anchoa tricolor* (Spix & Agassiz, 1829) along its ontogenetic development in an subtropical estuary at South America

Carvalho, B.M.; Volpedo, A.V.; Vaz-dos-Santos, A.M.; Spach, H.L.

Contact e-mail: bmaicarvalho@gmail.com

Abstract reference: IVE_Piñeiro_12

Seasonal climatic conditions influencing daily growth pattern on young of the year European hake (*Merluccius merluccius*) off NW Spanish waters

Piñeiro , C.; Rodríguez-Fernández, L.; Cabrero, A.; Nava, E.; García, A.

Contact e-mail: carmen.pineiro@vi.ieo.es

Abstract reference: IVE_Nuñez_15

Determination of larval growth stages in the codlet *Bregmaceros cantori* (Pisces: Bregmacerotidae) in Cariaco Trench, Venezuela

Núñez, J.G.; Marín, B.; Martínez, A. T.; Narváez, M.; Ariza, L.A.

Contact e-mail: jgnp31@gmail.com

Abstract reference: IVE_Vaz-dos-Santos_16

Do otoliths express the polyphasic growth of the main species in a coastal subtropical ecosystem?

Vaz-dos-Santos, A.M.; Gris, B.; Pereira-Junior, M.R.; Giombelli-da-Silva, A.; Arruda Justino, A.

Contact e-mail: andrevaz@ufpr.br

Abstract reference: IVE_Cisterna_17

Estimation of the age of jack mackerel (*Trachurus murphyi*) using daily growth rings in sagittae otoliths

Cisterna, L.; Arancibia, H.; Araya, M.

Contact e-mail: liliancisterna@udec.cl

Abstract reference: IVE_Frothingham_18

Exploring Arctic Cod *Boreogadus saida* life history from nearshore Beaufort Sea with the use of otolith microchemistry

Frothingham, A.; Norcross, B.

Contact e-mail: afrothingham@alaska.edu

Abstract reference: IVE_Katayama_19

Four types of otolith opaque zone

Katayama, S.; Gleadall, I.G.; Ito, K.

Contact e-mail: skata@m.tohoku.ac.jp

Abstract reference: IVE_Bernal_20

Growth patterns of the dominant lanternfish *Ceratospelus maderensis* (Pisces: Myctophidae) from the western Mediterranean

Bernal*, A.; Real*, E.; Olivar, M.P.; Molí, B.; Morales-Nin, B.

* Equal author roles

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Abstract reference: IVE_Valero-Rodriguez_21

Identifying seasonal patterns on daily growth of meagre otoliths

Valero-Rodriguez, J.M.; Gil, M.M.; Palmer, M.; Arechavala-Lopez, P.; Pérez-Mayol, S.; Sanchez-Jerez, P.; Morales-Nin, B.

Contact e-mail: jmvvr@alu.ua.es

Abstract reference: IVE_Manaja_22

Influence of temperature and fish size on otolith growth of laboratory-reared Atlantic cod (*Gadus morhua* L.) juveniles

Manaja, R.H.; **Folkvord, A.**; Geffen, A.J.

Contact e-mail: rmanaja@kfupm.edu.sa

Abstract reference: IVE_Costa_23

Intraspecific variation in the shape of the asteriscus otoliths of *Prochilodus nigricans* in three rivers of the Amazon Basin, Brazil

Costa, R.M.R.; Amadio, S.A.; **Fabré, N.N.**

Contact e-mail: rosa_rcosta@yahoo.com.br

Abstract reference: IVE_Muntoni_24

Lapilli vs sagittae: results from the comparison of settlers of the striped red mullet

Muntoni, M., Frongia, C., Rocklin, D, Lambiase, E, Raventos Klein, N, García-Charton, J.A, D'Anna, G, Murenu, M.

Contact e-mail: ma.muntoni3@studenti.unica.it

Abstract reference: IVE_Bellodi_25

Life history parameters of the small Mediterranean-endemic skate, *Raja polystigma* Regan 1923, from Sardinian seas

Bellodi, A.; Cau, Al.; Marongiu, M.F.; Mulas, A.; Porcu, C.; Vittori, S.; Follesa, M.C.

Contact e-mail: abellodi@unica.it

Abstract reference: IVE_Odelstrom_26

Life-history/age structure of three-spined sticklebacks in the Baltic Sea

Odelström, A.; Ångström, C.; Olsson, J.; Bergström, U.

Contact e-mail: anne.odelstrom@slu.se

Abstract reference: IVE_Al-Anbouri_27

Otolith Based Age and Growth Studies on the Indian Oil Sardine, *Sardinella longiceps* Valenciennes, 1847 From Muscat, Sultanate of Oman.

Al-Anbouri, I.

Contact e-mail: camry2005@yahoo.com

Abstract reference: IVE_Chung_28

Otolith microchemistry reveals ontogenetic records of vertical migration and metabolism of four dominant deep-sea fish in the Northeast Atlantic

Chung, M-T.; Shores, D.; Trueman, C.N.

Contact e-mail: mc7e10@soton.ac.uk

Abstract reference: IVE_Murie_32

Spatial and Temporal Effects of the Deepwater Horizon Oil Spill on Growth of Spotted Seatrout and Red Drum in the Gulf of Mexico

Murie, D.; Parkyn, D.

Contact e-mail: dmurie@ufl.edu

Abstract reference: IVE_Fotiadis_33

The thickness of daily rings statoliths of *Todaropsis eblanae* and the relationship between the month, age and sex

Fotiadis, N.

Contact e-mail: arcanum2009@hotmail.com

Abstract reference: IVE_Albuquerque_34

Tracking the use of freshwater streams by juvenile mullets (*Mugil liza*) through the analysis of Sr:Ca ratios in otoliths

Albuquerque, C.; Oliveira, M.C.L.M.; Conдини, M.V.; Garcia, A.M.

Contact e-mail: doccqa@yahoo.com.br

Abstract reference: IVE_Lill_35

Two-spot PIXE analyses of polished otoliths for identification of anadromous whitefish in the Baltic Sea

Lill, J-O.; Himberg, M.; Slotte, J.M.K.; Heimbrand, Y.; Florin A-B.; Hägerstrand H.

Contact e-mail: jlill@abo.fi

Abstract reference: IVE_Prokhorova_37

Using of individual biological features as auxiliary indicators in the age reading

Prokhorova, T.

Contact e-mail: alice@pinro.ru

Abstract reference: IVE_Pérez-Mayol_38

Were you a reared meagre? Marking of *Argyrosomus regius* otoliths to identify recaptures

Pérez-Mayol, S.; Gil, M.M.; Grau, A.; Morales-Nin, B.

Contact e-mail: silvia@imedea.uib-csic.es

Slideshow Poster Session (IVSS)

Abstract reference: IVSS_Chang_01

Age and growth of *Chrysochir aureus* and *Otolithes ruber* in the southwestern water of Taiwan

Wang, C.W.; Wang, L.P.; Chen, M.H.; Chao, N.L.; Chang, C.W.

Contact e-mail: changcw@nmmba.gov.tw

Abstract reference: IVSS_Neil_02

Age validation of NZ Puaa: the relationship between age and growth rings on the shell

Neil, H.; Naylor, R.

Contact e-mail: helen.neil@niwa.co.nz

Abstract reference: IVSS_Loh_03

Otolith atlas of Malaysia

Loh, K-H.; Sasekumar, V-C.C.A.; Dhillon, S.K.; Chang, C-W.

Contact e-mail: khloh@um.edu.my

Abstract reference: IVSS_Zitek_04

Transgenerational marking of freshwater fish otoliths using enriched stable Sr isotopes

Zitek, A.; Irrgeher, J.; Prohaska, T.

Contact e-mail: andreas.zitek@boku.ac.at

Abstract reference: IVSS_Piñeiro_05

How do otoliths grow? Linear daily growth increments and areas calculated in single-labeled otoliths from oxytetracycline-injected hake (*Merluccius merluccius*)

Piñeiro, C.; Rey, J.; Rodriguez, L.; Goñi, R.; Gomez, R.

Contact e-mail: carmen.pineiro@vi.ieo.es

Age Validation Workshop Posters

Abstract reference: WSAgePHastie_12

Upwelling impacts on a California Current bomb radiocarbon reference chronology and petrale sole (*Eopsetta jordani*) age validation

Hastie, J.

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Corroboration of faster annual growth rate of black anglerfish (*Lophius budegassa*) in Spanish Mediterranean based on length frequency analysis.

Barcala, E.

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Comparison of otolith and scale readings for age estimation of common dentex (*Dentex dentex*)

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Daily rings observations in blackspot seabream *Pagellus bogaraveo* (Brünnich, 1768)

Bekas, P.; Mytilineou, Ch.

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Comparing two methods of age determination of herring (*Clupea harengus*) in the Bothnian Sea and the Bothnian Bay

Blass, M.; Eklund, J.; Elfving, M.; Heimbrand, Y.; Jernberg, C.; Kaljuste, M.; Odelström, A.

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Age validation of the jack mackerel (*Trachurus murphyi*) off Chile

Cerna, F.; Campana, S.; Moyano, G.; Bocic' V.; Ojeda, V.

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Using otolith weight to improve age estimation in red mullet *Mullus barbatus*

Chatzisyrou, A.; Anastasopoulou, A.; Mytilineou, Ch.; Bekas P.; Kallianiotis, A.; Haralabous, J.

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Validation of daily increments and a marine entry check in juvenile Sockeye Salmon using experimental rearing and microchemical techniques

Freshwater, C.

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Clarity and accuracy of ageing transverse sectioned otoliths from black sea bass, *Centropristis striata*, from the Northwest Atlantic Ocean

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Estimation of age and growth of juveniles of two European anglerfishes, *Lophius budegassa* and *L. piscatorius*, in the north-eastern Atlantic waters, from otolith microstructure analysis.

Hernández, C.; Landa, J.; Barrado, J.; Antolínez, A.; Villamor, B.; Navarro, M.R.

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Multicriteria approach for validating the first winter ring deposition in Eastern North Sea plaice (*Pleuronectes platessa*) otolith: preliminary study

Johansson, J.; Bland, B.; Vitale, F.; Carbonara, P.

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Preparation of hake *Merluccius* spp. otoliths for routine annual age determination in Namibia

Kashava, S.; Paulus, S.C.; Wilhelm, M.R.

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Potential use of otolith weight to estimate stripped mullet *Mullus surmuletus* age

Kousteni, V., Denaxa, M., Anastasopoulou, A., Bekas, P., Haralabous, J., Mytilineou, Ch.

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Lampri, P-N.; Bekas, P.; Mytilineou, Ch.

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Precision in age estimates in Octopus vulgaris stylet increments analysis

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Age and growth of Atlantic bluefin tuna using otolith microstructure and macrostructure: The case study of the Mediterranean fish

Megalofonou, P. and Dean, J.M.

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Montanini, S.; Vallisneri, M.; Pérez-Mayol, S.; Palmer, M.; Morales-Nin, B.

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Application of quality control ageing of *Trachurus murphyi* and *Merluccius gayi gayi* through procedures approved by ISO 17025

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Assessing the otolith weight based ageing in picarel *Spicara smaris* (*Osteichthyes:Centracanthidae*)

Mytilineou, Ch., Karkani, M., Bekas, P., Tziertzidis, D., Haralabous, J.

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Pattoura, P.; Mytilineou, Ch.; Lefkaditou, E.; Haralabous, J.

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Enhancing otolith microstructure by a plastic inclusion technique

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stimating the age and growth of European eel (*Anguilla anguilla*) in Estonian lakes using „buring and cracking“ method on otoliths

Silm, M.; Järvalt, A; Bernotas, P.; Mäe, A.

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Otolith ageing and preliminary validation of *Lampanyctus crocodilus* (Risso, 1810) in the North-western Ionian sea (Central Mediterranean)

Sion, L.; Indennitate, A.; Carlucci, R.; Carbonara, P.; D'Onghia, G.

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Age Validation of lemon sole (*Microstomus kitt*), using marginal increment analysis

Smith, J.

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Description of the main sources of error associate with the age allocation of Chilean jack mackerel *Trachurus murphy* (Nichols, 1920)

Valero, C.; Muñoz, L.; Cerna, F.

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Abstracts by Theme Session

Theme I: Environmental Indicators

Plenary session (IA)

Abstract reference: IA_Vasconcelos_Key

Calcified tissues as environmental indicators

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The difficulty in measuring changes in ecosystems resulting from natural and anthropogenic drivers has encouraged the development of indicators. Moreover there is an increasing demand for widely applicable indicators, providing tools both to scientists and policy-makers. Environmental indicators are measures of biological, physical or chemical components that reflect the state or changes in the environment. The capacity of calcified tissues (fish otoliths, scales and bones, mollusk shells, and corals) as environmental indicators has been tested in multiple ways during the last decades. Focus has been directed chiefly to their use as temporally resolved tracers/records of environmental conditions, *i.e.* to the relationship between properties of calcified tissues (*e.g.* elemental and isotopic composition; otolith shape, asymmetry, matrix opacity; scale and bone deformation; growth rates) and environmental conditions (*e.g.* element and isotope input and availability, temperature, salinity, organic matter sources, ocean acidification). In addition to their applicability as environmental indicators of current or recent states of the environment, attention has also been given to the use of calcified tissues from fossils and long-lived organisms as records of historic times. Despite notable advances, the current degree of uncertainty in many of these approaches is high and hinders their broad applicability, and research directed at validation is urgent. Ultimately these properties are not dissociable from the use of otoliths as individual, population and community indicators.

Abstract reference: IA_Daverat_00

Protolithe: protoscape and fish odyssey

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The installation « Protolithe » was created as an experience of science, literature, video and music, to immerge the public into an aquatic atmosphere where the mysteries of the journeys of eels, shads and flounders are revealed through art and science. This collaborative initiative between a poet, an analytical chemist, a fish ecologist and a music composer, is based on the information retrieved from fish otoliths. Protolithe is a collection of poems, haïkus, questioning the epic journeys of migratory fish. The installation is based on the results and tools used for collaborative

research projects between ecology of diadromous fish and laser ablation analytical chemistry. Protolithe is composed of two parts. The first part illustrates the geographic milestones of migrations, embodied by water steles, made of glass tubes filled with water from the different habitats encountered by the diadromous fish species and engraved with the collection of poems “protoscape”. The second part of the installation is “Odyssey”, a collection of poems engraved in microscopic letters on otoliths, after the story of each individual fish. The installation is completed with a video of fish passage to picture the hazards of migration on the background and an immersive music. Protolithe installation was inspired by the fish individual migratory patterns retrieved from otolith microchemistry of the presented otoliths, formerly used in different projects addressing European eel, flounder and Shad individual diversity of migration pathways. Challenging femtosecond laser ablation techniques were performed to both analyse specific life stages of fish, and engrave the poems in 30 µm height letters.

Abstract reference: IA_Gillanders_01

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Using fish ear bones as proxies for tracing changes in climate

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The world is seeing unprecedented changes in climate. Increased anthropogenic CO₂ emissions associated with industrialization have resulted in an increase in atmospheric CO₂ over the past two centuries. Associated with this atmospheric CO₂ increase has been an increase in oceanic CO₂ levels leading to a decrease in both pH and carbonate ion concentration, termed ocean acidification. Tracing either changes in pH through time or the pH history of fish throughout their life is difficult, but the ear bones (otoliths) of fish, which grow incrementally and accrete CaCO₃ along with trace elements may provide a novel option. Our aim was to determine if trace elements in the otoliths of fish could be used as a natural tracer of variation in pH. We used two approaches to test this aim, a laboratory experiment and examination of field-caught fish that were collected from naturally elevated CO₂ conditions. Our controlled laboratory experiment exposed fish to current and a future level of pH in combination with increased temperature. Benthic site-attached fish were also collected from two shallow water CO₂ vent sites with collections being undertaken at varying levels of CO₂. Trace elements in otoliths from both the laboratory and natural experiment were analysed using laser ablation inductively coupled plasma-mass spectrometry (LA ICP-MS). Relationships between CO₂ and trace elements will be presented. This research helps to understand how changing environmental conditions are reflected in fish otoliths which has important implications for monitoring fish movement.

Abstract reference: IA_Grammer_02

Investigating the bomb radiocarbon flux in the southern Pacific Ocean with otolith radiocarbon

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Otoliths (ear stones) from long-lived fish provide an indirect method to examine levels of radiocarbon (¹⁴C) resulting from nuclear weapons testing in the 1950s and 1960s (bomb radiocarbon, $\Delta^{14}\text{C}$), thereby allowing the changes in distribution of ¹⁴C in a region to be interpreted. Our research aims to examine the flux of the “bomb pulse” through the water column from surface waters to depths ≤ 1000 m in the south-western Pacific Ocean purely by using otolith radiocarbon. We examine ¹⁴C series from marine surface waters (Australasian snapper, *Chrysophrys auratus*; nannygai, *Centroberyx affinis*), 400 - 500 m (ocean perch, *Helicolenus barathri*) and 800 - 1000 m (orange roughy, *Hoplostethus atlanticus*), all collected from south-western Pacific areas. We compare ¹⁴C measurements micro-sampled from ocean perch otoliths to otolith surface water reference values and deep-water ¹⁴C values from orange roughy to establish for the first time a mid-water ¹⁴C series for depths of $\sim 400 - 500$ m through the years of 1947 - 2009. The linear trends of $\Delta^{14}\text{C}$ during the greatest periods of increase for the three time series were correlated. For all series comparisons, the intercepts of increase differed, but the slopes were different only between the surface and deep-water series. These results indicate a 5 - 10 year lag between the surface and depths of 400 - 500 m and a 10 - 20 year lag between the surface and depths of 800 - 1000 m with depression of the slope of ¹⁴C increase as depth increases. Overall, application of fish otoliths to examine the bomb pulse of ¹⁴C provided valuable insights into the timing of ¹⁴C flux into depths approaching 1000 m in the south-western Pacific Ocean, an area deficient in instrumental temporal ¹⁴C records in waters below the mixed layer.

Abstract reference: IA_Pecheyran_03

Virtual beam shaping femtosecond laser ablation: a new tool for sensitive high spatial resolution otolith microchemistry

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Trace element analysis in fish otoliths has received a growing interest in the last decade. Due to significant advances in laser microsampling coupled to inductively coupled plasma mass spectrometry (LA/ICPMS) trace elements are now commonly used to determine migration patterns or as a fingerprint of natal origins and habitats. However some constraints related to the irregular structure of otoliths at the micron scale, the need for multielemental detection from pg/g to ug/g levels (sometimes associated to isotopic composition) and the lack of certified reference material are now limiting this technique. When high spatial resolution is required, small laser beams must be used which drastically limits the analytical sensitivity and hampers the determination of trace elements. Ultra short pulses of femtosecond lasers are now considered as a major advance for improving accuracy and precision. We have developed new analytical methodologies based high repetition rate (up to 100000 shots/s) femtosecond laser combined to fast galvanometric laser

beam movement: this approach allows virtual beam shaping particularly suitable for improving spatial resolution and analytical sensitivity. It is then possible to adapt the size and the shape of the laser beam to match perfectly with the sample morphology. We will show through several cases of study regarding multielemental detection and Sr isotopic composition the benefits of this new technique: elongated laser beams for transects of complete life history, corona laser beams to specifically record the first days of life, single spots for maternal signature or specific beam shapes perfectly matching the striae for the last days of life.

Abstract reference: IA_Shoji_04

Natural habitats contribute more to an estuarine fish production than artificial habitats do: variability in growth and mortality rates of larval and juvenile temperate bass cohorts estimated from otolith microstructures

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Mortality of larval temperate bass *Lateolabrax japonicus* cohorts during the period of post-migration into estuaries were compared between Ohta Drainage Channel (channel river: CR) and Temma River (natural river: NR) in western Seto Inland Sea, southwestern Japan. Since the two river mouths are located within 1 km, the larval habitats were exposed to the same pattern of larval supply and temporal fluctuations in freshwater flow and tide after migration into the estuarine habitats. However, during the period of high precipitation, freshwater discharges more abruptly and produces increases in CR through human control. In order to test the hypothesis that an increase in freshwater discharge affects larval mortality, cohort-specific growth (G, d⁻¹) and mortality (M, d⁻¹) coefficients and the ratio of G:M as a proxy of larval production were compared between the two estuaries. The G and M were estimated through 1) repeated sampling at fine time intervals (6-15 d) from February to May, 2) application of otolith daily increments for cohort identification and 3) standardization of abundance at age based on the length-dependent catch efficiency of the sampling gear in order to minimize the sampling error. The Gs ranged between 0.009 and 0.024 without significant difference between CR and NR. The Ms in CR (0.184-0.239) was significantly higher than those in NR (0.140-0.148), resulting in an average ratio of G: M (0.123) in NR, about double of that in CR (0.059). Temperature and prey concentration were not significantly different between the two estuaries throughout the sampling period, while decrease in salinity was more prominent in CR from March to April due to precipitation. Among the major sources of mortality in fish early life stages (starvation, physical processes and predation), physical processes including passive transportation due to increase in freshwater discharge seemed to have highly contributed to the inter-river difference in survival of the larval cohorts since differences in larval supply, prey availability and predation risk between the two estuaries were considered to be minimal.

Do variations in the elemental concentrations in otoliths of marine fishes really track variations in water chemistry?

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Otolith microchemistry has proven its use in stock discrimination and movement studies. Indeed, otolith microchemistry has been a surprisingly sensitive tool for stock discrimination in marine settings where water composition is relatively homogenous. However, despite intense study, predictive relationships between water chemistry and otolith chemistry remain elusive. Many experimental studies have been performed usually with an end aim of calculating a partition or distribution coefficient -effectively the ratio between element concentration in water and in the otolith. The concept of the distribution coefficient hails from inorganic geochemistry, where equilibrium reaction processes dominate elemental uptake. Here we argue that this concept is not applicable (and indeed misleading) when applied to element incorporation into biominerals of higher organisms, where physiological variables influence, and may indeed dominate, variations in biomineral trace element composition. An alternative conceptual framework considers elemental concentrations as a consequence of differential kinetics of uptake and excretion in multiple body pools. Biokinetic models are commonly used in toxicology, and here we develop a simple biokinetic model and apply this to results from a long-term element partitioning study in plaice. We show that temporal variations in element / calcium ratios (especially Sr/Ca ratios) observed in blood and otolith tissues can be readily explained by kinetic processes associated with metal transport in body fluids, but do not fit an equilibrium uptake model. A biokinetic view of element uptake explains apparently contradictory observations of calculated distribution coefficients between water and otolith, and partially explains the surprising success of otolith microchemistry as a stock discrimination tool in marine settings. We argue that it is time to abandon distribution coefficients and the implied equilibrium concepts. Attempts to explain or predict variations in otolith trace element composition according to seawater chemistry are likely to fail. Instead, we suggest that by understanding processes that influence rates of element uptake and excretion in fishes, we may be able to unlock a wealth of physiological information contained in otolith trace element data.

Parallel Session IB (Anthropogenics effect, Geotags)

Abstract reference: IB_Limburg_01

Shades of Sophie Dove? The potential of eye lens chemistry as a complementary archive of environmentally relevant information

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Eye lenses are formed of the protein crystallin, and grow throughout an organism's life. The external cells are metabolically active whereas the interior cells are not, but remain in place and essentially isolated, *i.e.* not subject to metabolic re-working. We have been studying the feasibility of using eye lens chemistry as a complement to otolith chemistry. Eye lenses have very little calcium and strontium, thus easing the ability to detect other trace elements. We have produced 2-D maps of fish lens chemistry using scanning X-ray fluorescence microscopy at the Cornell synchrotron. We consistently detect bromine, copper, rubidium, selenium, and zinc across a wide range of fish taxa; and in some cases we detect mercury and other trace elements such as lead or gold. We will discuss the feasibility of using the chemical "information" archived in eye lenses as a complement to that archived in otoliths.

Abstract reference: IB_Tracey_03

Ongoing ageing of a habitat-forming deepsea coral *Solenosmilia variabilis* – information to assess recovery potential from anthropogenic impacts

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Some coldwater corals can form large reef-like three-D matrix structures in the deepsea that provide habitat for numerous other species. These structures are often fragile, and easily damaged by human activities such as bottom trawling. Our knowledge of the potential recovery rate of these habitats and their associated communities is growing but limited by our lack of knowledge on the age and growth of the main reef-forming coral species. One of the most widespread species in the South Pacific is *Solenosmilia variabilis*. In a preliminary study the age of this coral species was assessed in New Zealand for the first time, using radiocarbon content ($\Delta^{14}\text{C}$). Linear growth rates ranged from 0.3 to 1.3 mm/yr. Using a conservative estimate of matrix height of ~20cm, it could take hundreds of years (~150-660) for a colony to attain this height, or ~380-1700 years to build a diameter of 1 meter. Our current research is to build on our initial estimates and radiocarbon date the long dead, recently dead, and live stony coral colonies of the coral matrices to progress our understanding of their living distribution. Our defining of the age, growth and restoration timeframes of deep-sea coral matrices in the region will inform management of deep-sea habitats around New Zealand.

Abstract reference: IB_Clemmensen_04

Effects of ocean acidification on otolith growth and size-selective mortality of larval Atlantic cod (*Gadus morhua* L.) and Atlantic herring (*Clupea harengus* L.)

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The effects of ocean acidification on otolith growth and on size-selective mortality were investigated by rearing Atlantic cod (*Gadus morhua* L.) and Atlantic herring (*Clupea harengus* L.) in three pCO₂ concentrations, control-370, medium-1800, and high-4200 ppm in outdoor mesocosms at Espesgrend Station, Norway in Spring 2010 and under laboratory conditions at control-400 and low-1000 ppm pCO₂ concentrations in Kristineberg, Sweden in Spring 2013. Hypercalcification of otoliths was observed from 7 to 46-dph on cod larvae cultured at elevated pCO₂ concentrations in the mesocosms. The *sagittae* and *lapilli* were usually largest at the high pCO₂ treatment followed by medium and control treatments. The biggest difference in mean otolith surface area (normalized with fish length) was with *sagittae* at 11-dph with medium and high treatments being 46% and 43%, respectively, larger than the control group. Significant increase in otolith surface area was also found in cod larvae under laboratory settings at 1000 ppm. In contrast, herring larvae exposed to elevated pCO₂ concentrations produced significantly smaller otoliths (both *sagittae* and *lapillae*) relative to the control group at 32 and 39-dph. The largest difference (33%) in otolith area was between the *sagittae* of control and high treatments at 32-dph. The possible mechanisms of the differences in the responses of net otolith growth between cod and herring larvae will be discussed. Repeated samplings within the same population allow for an accurate determination of size-selective mortality by comparison of otolith size-at-age based on samples from different dates and back calculations. Using otolith microstructure analysis we provide examples to evaluate mortality patterns in fish larvae being treated with different levels of CO₂ during their larval development. This will allow determining whether survival in response to CO₂ treatment is random or size-selected. The analysis will give new insights into the “characteristics of the survivors” after CO₂ exposure.

Abstract reference: IB_Granneman_05

Detecting interannual shifts in otolith microchemistry before, during, and after the Deepwater Horizon oil spill

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The objective of this study was to describe the broad timescale otolith element composition of several Gulf of Mexico fish species from otolith regions that correspond to periods prior to, during,

and after the Deepwater Horizon (DWH) oil spill in April 2010. We analyzed otoliths of the following fish species collected from 2011 to 2013 in the Gulf of Mexico: *Lutjanus campechanus*, *Epinephelus morio*, *Epinephelus flavolimbatus*, *Pagrus pagrus*, *Urophycis floridana*, and *Lopholatilus chamaeleonticeps*. Otoliths were analyzed using laser-ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) for a suite of 26 isotopes: ^7Li , ^{23}Na , ^{24}Mg , ^{31}P , ^{45}Sc , ^{51}V , ^{53}Cr , ^{55}Mn , ^{57}Fe , ^{59}Co , ^{60}Ni , ^{63}Cu , ^{64}Zn , ^{65}Cu , ^{72}Ge , ^{85}Rb , ^{88}Sr , ^{89}Y , ^{114}Cd , ^{118}Sn , ^{137}Ba , ^{197}Au , ^{208}Pb , ^{232}Th , and ^{238}U , with ^{43}Ca used as an internal standard. We ablated otoliths along a transect that extended from the primordium to the edge of the otolith, providing element profiles spanning the entire life of the individual. This technique allowed us to establish baseline otolith microchemistry measurements for fish prior to the DWH oil spill. Using canonical analysis of principal coordinates (CAP), we found significant differences in otolith microchemistry among time periods (before, during, and after the spill) for all of the species tested. We observed that the concentrations of several elements closely associated with salinity (^{23}Na , ^{55}Mn , and ^{137}Ba) changed significantly during these time periods. This may have been due to the intentional increase of Mississippi river discharge during the DWH oil spill to displace surface oil away from the coast of Louisiana. Additionally, a significant increase in ^{63}Cu , ^{65}Cu , ^{64}Zn , and ^{118}Sn was observed for most species both during and after the oil spill. Thus, applying broad timescale methods to this dataset revealed a shift in otolith microchemistry coincident with the time period of the DWH oil spill for all species in the study.

Abstract reference: IB_Yang_06

An investigation of connectivity among estuarine tapertail anchovies *Coilia nasus* from the Yangtze River, Yellow Sea and Poyang Lake

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Estuarine tapertail anchovy (*Coilia nasus*) is an anadromous species, and adult *C. nasus* migrates to the Yangtze River for spawning from sea areas in early February to the end of April. *C. nasus* is a highly commercial and valuable species in China due to its delicacy and traditional consumption culture. The anchovy from the Yangtze River is probably the most expensive fish in the world. The maximum price even reached as high as about \$1000/kg, and \$9600 for a single fish with 45.3 cm TL and 0.325 kg BW in Jiangsu Province in 2012. Because of empirical limitations with the current methods of catch-survey analysis, it remains difficult to effectively understand the exact location of spawning sites, the diadromous migration features of this species, and the relationship between the anchovy stocks among the Yangtze River, and its connected lakes and adjacent seas. In the present study, we collected *C. nasus* anchovies from mainstream and estuary of the Yangtze River, river-connected lakes (the Taihu Lake and Poyang Lake), and estuary adjacent seas (the Yellow Sea and East China Sea). Their otolith Sr and Ca was analyzed by EPMA to estimate the habitat history. At the same time, body morphology and otolith microstructure were investigated for the purpose of mutual corroboration. Our results suggest, for the first time, that *C. nasus* migrates anadromously for spawning into the Poyang Lake, which locates more than 800 km upstream of the Yangtze River mouth. Furthermore, a relatively close connectivity may exist between the *C. nasus* anchovies of the Yangtze River and the adjacent Yellow Sea. *C. nasus* grows in the Yangtze River nearly for one year, and then enters the brackish and sea water habitats which can be a nearly straight line for even as far as 300 km off the coastline.

Abstract reference: IB_Vignon_07

Extracting environmental histories from sclerochronological structures - Recursive partitioning as a mean to explore multielemental composition of fish otolith

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During the last decades, ecologists have paid a great deal of attention to the proper techniques to track fish movement in the wild. In this context, otoliths have proved useful and became increasingly used in a wide range of species worldwide. Otolith material incorporates elements from the surrounding water in a layered manner that preserves the timing of deposition. Elemental composition along otolith centre–edge transects thus represent a permanent record of the growing habitats experienced by the fish throughout its entire life time. While the analysis of these elements is of primary importance in fishery management and population ecology, the traditional approaches mostly fail to capture the multivariate nature of structural change in time series data. Here, I propose a simple approach aiming at coping with the multi-elemental compositional otolith transect in a multi-scale quantitative manner. More specifically, the proposed recursive partitioning method try to bias the chronological clustering process by accommodating some form of user-specified constraints based on both intra- and inter-groups characteristics. This allows researchers to focus on biologically relevant information (*i.e.* the one researcher look for and can interpret). Chronological clustering based on recursive partition is easily interpretable and directly applicable to detection of hidden discontinuities in any multivariate time series. In a multi-proxy context, it also provides a powerful exploratory tool for assessing the relative importance of cross-correlated variables in structuring the environmental histories, as well as the congruency between variables. Ultimately, approach demonstrates the actual interest in using advanced processing techniques to fully exploit the rich potential of individual biological archives, such as fish otoliths, to characterize individual life history. This appears as a promising new tool for ecologists wishing to extract environmental histories sclerochronological structures and would be a great help for practitioners working with chemical fingerprints.

Abstract reference: IB_Yamashita_08

Relative contribution of estuarine nursery areas to the adult population of the temperate seabass *Lateolabrax japonicus* in Tango Bay, as revealed by otolith Sr:Ca ratios

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The temperate seabass *Lateolabrax japonicus* is a euryhaline species found along northeastern Asian coasts. Landings of this species from Japanese coastal waters have been increasing over the past 25 years. Juveniles utilise various coastal habitats as nurseries, such as eelgrass beds, sandy beaches, tidal flats and estuaries. The importance of estuaries as nursery areas for temperate

seabass has been emphasised by several recent studies. To assess the values of estuarine nursery grounds, the contribution of estuarine areas to the production of temperate seabass populations was estimated using otolith Sr:Ca ratios in Tango Bay, Sea of Japan. Otolith Sr:Ca ratios of juveniles collected from freshwater and brackish areas of the Yura River estuary, which flows into Tango Bay and coastal areas were analysed to determine a criterion for distinguishing whether individuals used estuarine or coastal areas as nurseries. The mean value of Sr:Ca ratios at the edges of otoliths of juveniles showed significant differences between areas ($P < 0.01$), measuring 4.9×10^{-3} , 4.0×10^{-3} and 2.4×10^{-3} in the coastal, estuarine and freshwater areas, respectively. The mean minus standard deviation of the Sr:Ca ratio of individuals from the coastal area was 4.4×10^{-3} and this value was used to estimate the nursery type utilised by juveniles. Otolith Sr:Ca ratios of adult seabass collected around Tango Bay were also analysed. Thirty-nine of 107 (36 %) adult seabass were estimated to have used the estuary as a nursery during their juvenile stage according to their Sr:Ca chronologies. Despite the small area covered by estuarine nurseries compared with coastal waters, the estuary contributed considerably to the adult seabass population. This indicates the estuary plays an important role as a nursery area for seabass juveniles.

Abstract reference: IB_Sih_09

Near-reef elemental signals in the otoliths of *Pomacentrus amboinensis* (Pomacentridae)

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Pre-settlement processes that determine where and when a larval coral-reef fish will settle can have important implications not only individual survival, but also population distributions and abundances. However, many key factors, such as how long returning larvae spend close to reefs prior to settlement, are poorly understood. This study combined field and laboratory studies, coupled with laser ablation inductively-coupled plasma mass spectrometry (LA-ICP-MS) and otolith microstructure analysis (daily increments and settlement marks) to determine the length of time larval fish spent near a reef prior to settlement. The otoliths of Ambon damselfish, *Pomacentrus amboinensis*, collected from multiple neighbouring reefs in the southern Great Barrier Reef, showed clear differences in their elemental signatures prior to, and following, settlement. However, individual profiles also revealed substantial variation, with an increased otolith Ba:Ca ratio (near-reef signature) occurring right at settlement in some individuals, but up to 8 days prior to settlement in others. Increment widths, often used as a proxy for growth, decreased approaching the settlement mark. Differences in otolith elemental chemistry were also found between fish kept in Lagoon (reefal) and Ocean (inter-reefal) waters, however, these differences did not reflect the highly variable elemental composition of water sampled from each location. Poor discrimination of multi-element signatures was observed among fish from different reefs pre-settlement, but discrimination improved in the post-settlement phases of otoliths. This study demonstrated clear and consistent near-reef elemental signatures in fish around the time of settlement. We suggest these differences are due to a combination of water chemistry and physiological influences (i.e. feeding). Combining LA-ICP-MS with otolith microstructure analysis can provide high resolution information on the early life history of reef fishes.

Parallel Session IC (Chronology, Climate and Growth)

Abstract reference: IC_Swearer_01

A stitch in time: using chemical chronologies from otoliths to reconstruct larval dispersal environments

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Otolith elemental profiles of larval fishes have great potential to provide insight into the environmental conditions experienced during larval development, particularly in areas which experience spatial gradients or heterogeneity in physico-chemical properties of water. Here we explore stitching otolith elemental profiles from larval fish to generate longer time series of environmental histories to assess the extent to which otoliths record changes in water mass properties in bay versus open coast pelagic environments during the larval period in a temperate reef fish. Using dynamic regression models, we show that high-resolution time series of larval otolith chemistry do reflect changes in local environmental conditions, providing a new approach for shedding light on the dispersal ecology of fishes.

Abstract reference: IC_Power_02

Otolith-derived estimates of marine temperature use by West Greenland Atlantic salmon (*Salmo salar*)

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Otolith-derived estimates of mean marine temperatures experienced by one-sea-winter (1SW) Atlantic salmon (*Salmo salar*) of North American origin were determined for fish sampled off West Greenland in 2009 and 2010. Otolith material from the edge corresponding to the second summer at sea was sub-sampled, via micro-milling, and analysed by mass spectrometry to produce stable oxygen isotope ($\delta^{18}\text{O}$) values. Retrospective estimates of mean temperatures experienced were calculated for each fish for the second summer growth period from otolith and seawater $\delta^{18}\text{O}$ values using a salmonid-based fractionation equation. Temperature estimates did not differ significantly by summer period (early versus late) or capture year, although there was considerable among-individual variability. Comparison of otolith-derived temperature estimates with available sea surface temperature (SST) data for the Labrador Sea for the same summer growing season period indicated that the otolith estimated temperatures corresponded well with mean SST for the region. Otolith-derived temperature estimates also broadly agreed with data on marine

temperature use published in the literature or derived from the limited number of data storage tags that have been retrieved for Atlantic salmon, with 80 percent of all individuals using temperatures in the 4.7 to 10.9°C range. Among-individual differences in temperature use did not correlate with measured otolith growth zone widths and there was no relationship between the otolith growth zone width and gonad weight. The among-individual variability in temperature use was interpreted to reflect differential use of the environment possibly driven by differences in opportunistic feeding behaviour known to predominate among Atlantic salmon but could also reflect variable thermal preferences of salmon from different stock origins.

Abstract reference: IC_Helser_03

A 200 year archeozoological analysis of Pacific cod life history as revealed through Ion Microprobe oxygen isotope ratios in otoliths

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Fish otolith oxygen isotope ratios ($\delta^{18}\text{O}$) are considered “flight recorders,” providing records of sea water temperature and habitat use over the animal’s life span.

We measured $\delta^{18}\text{O}$ values in modern and archeological Pacific cod otoliths using a high precision ion microprobe. Values of $\delta^{18}\text{O}$ were measured in as many as eighty 10-micron spots along transects from the otolith core to its margin with high spot-to-spot analytical precision ($\delta^{18}\text{O} \pm 0.3\%$). We obtained sample densities along a linear transect that were at least 2 to 3 times greater than micromilling/conventional mass spectrometry techniques. From modern Pacific cod otoliths (using *in situ* temperatures from electronic archive tags) we calibrated the fractionation equation of aragonite ($r^2 = 0.75$, $p < 0.001$, $\delta^{18}\text{O}_A = 2.13 - 0.25T^\circ\text{C}$) to predict sea water temperature. Sinuous variability of $\delta^{18}\text{O}$ values along core-to-margin transects likely reflect seasonal temperature changes and suggest similar longevity between modern and archeological cod. Generally increasing $\delta^{18}\text{O}$ values from the otolith core to the margin revealed an ontogenetic migration from warmer near shore habitat during the first year of life to cooler deeper waters at later ages, a behavior that has not changed over the past 200 years. A decline in the average $\delta^{18}\text{O}$ of core spot samples from archeological (200+, 100+ YBP) to modern otoliths suggest increasing sea surface temperatures from the late Little Ice Age to present. Temperatures calculated from the $\delta^{18}\text{O}$ in aragonite suggest a 2-3°C rise in coastal marine sea surface temperatures in the Gulf of Alaska over the last 200 years.

Abstract reference: IC_Wilhelm_04

Alaskan lake trout biochronologies as long-term indicators of climate and productivity

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High latitude lake ecosystems are likely to experience rapid changes in response to anthropogenic global warming with consequences for such processes as organismal growth rates, timing of seasonal events (phenology), species interactions, and overall levels of productivity. Establishing baseline climate-biology relationships is critical to benchmark these long-term changes, yet the biological time series necessary to do so are rare in these arctic and sub-arctic regions. To address this issue, we apply tree-ring analysis (dendrochronology) techniques to develop biochronologies from the annual growth increment widths of Alaskan lake trout (*Salvelinus namaycush*) otoliths. Samples were collected from lakes toward the northern and southern extremes of the species' range in Alaska, spanning approximately 60°N through 68°N. Final lake trout chronologies were exactly dated, multiple decades in length, and annually resolved (one value per year). The northern chronology strongly and positively ($p < 0.001$) correlated to temperature with peak relationships occurring in late summer. By contrast, a chronology developed from a network of southern lakes contained a lower degree of synchrony among individuals and was negatively ($p < 0.05$), and weakly, related to temperature. Indices of drought and precipitation were not significantly related to either chronology. Thus, warm temperatures appear to strongly facilitate growth at the northern limit of the species' range, but weakly limit growth at the southern limit of its range. Temperature is also related to tree-ring chronologies in the region, underscoring the sensitivity of fish and tree growth to warming as well as the complexity of biological response within and across diverse species and ecosystems.

Abstract reference: IC_Neil_05

Insights into historical New Zealand marine shelf productivity using ancient fish otoliths

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Determining the impacts of humans on New Zealand shelf ecosystems depends in part on estimation of the productivity of the ancient marine ecosystems compared with those of today. While in the modern period extensive collection of environmental and ecological data allows the cascading impacts of bottom-up, climate-forcing effects to be documented, other measures are required for the pre-instrumentation period. The hard parts of organisms can provide sufficient information to infer the interaction of environmental drivers and ecological responses. This paper details the evidence for changes in marine productivity contained within the bones, specifically the

ear bones or otoliths, of individuals of two species of fish, snapper (*Pagrus auratus*) and red cod (*Pseudophycis bachus*), caught by ancient Māori and preserved in their middens, and caught from the modern stocks. We used otolith incremental growth analysis and oxygen and carbon stable isotope analysis to infer the growth and environmental conditions experienced by fish in two regions (Hauraki Gulf and the Otago-Catlins shelf) over two periods; ~AD 1450 and the 20th century. Our results for both snapper and red cod suggest there is no difference between modern and midden otoliths, at the geographic sites studied, with respect to growth (juvenile increment or total otolith), temperature at age, migration at age, diet or/and metabolic rate, age of maturity, and total change in metabolic rate over the lifetime. We conclude that there is no discernable oceanic climatic regime or productivity difference recorded in otoliths from the Hauraki Gulf and Otago-Catlins regions around ~AD 1450 and the 20th century. This correlates well with atmospheric proxy data collated and interpreted for another part of the project that suggest that the climate in the two periods was very similar (although they are separated by a period of cool wet climate).

Abstract reference: IC_Ong_06

Contrasting environmental drivers of adult and juvenile growth in a marine fish

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Many marine fishes have life history strategies that involve ontogenetic changes in the use of coastal habitats. As the effects of climate change are likely to vary among habitats, this could potentially alter the balance between selective forces (predation and foraging) that drive sequential patterns of habitat occupancy. This is of particular concern in tropical Asia and Africa where such fishes form a major part of food resources for developing nations. We used a dendrochronology approach to examine the physical and biological drivers of adult and juvenile growth of mangrove jack (*Lutjanus argentimaculatus*) from north-western Australia. This species is a prized target of commercial and recreational fisheries and has juveniles that inhabit estuarine environments and an adult stage that resides on coastal reefs. For adults, a large-scale El Niño-Southern Oscillation (ENSO) process, the Niño-4 index, had the highest correlation with growth chronologies, highlighting the importance of atmospheric and oceanographic phenomena that operate at ocean-basin scales as determinants of growth patterns in adult habitats. Conversely, a different set of physical variables were correlated with the growth of juveniles inhabiting estuarine environments, notably local factors such as wind speed and rainfall. Our results show that the sensitivity to climate change of each life history stage of this species is likely to be very different, with implications for resilience and management of populations.

Abstract reference: IC_Davies Coad_07

Examining the interactions of growth, climate and recruitment of boarfish (*Capros aper*) for a better understanding of the recent population expansion

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The interannual growth patterns observed from otoliths of boarfish were examined from two geographically separate areas in the Northeast Atlantic. Given the observed increase in stock size and northward expansion of this species we chose an area “north” at the northern distribution range west of Ireland and “south” on the main fishing grounds south of Ireland. We investigated whether temperature changes influence the growth and recruitment dynamics of this species. Growth patterns were similar between the two areas with distinct years of faster and slower growth. The relationship between adult growth and temperature in the “north” was not significant whereas in the “south” the relationship was positive up to approximately 16 °C and growth rates suppressed in the years with temperature above that. Recruitment was positively correlated with adult growth the previous year and the relationships with various recruitment indices suggest spatial connectivity between the Celtic Sea and the Bay of Biscay. The age distribution in the two areas are similar and although boarfish can reach a maximum age of >30 years the younger age classes dominate. The observed stock expansion is therefore likely to be a consequence of increased recruitment under climatic conditions preferable to the physiological capabilities of the species.

Abstract reference: IC_Doubleday_08

Broad and local scale drivers of growth of an estuarine fish species and implications for climate change

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The analysis of annual growth increment patterns or ‘growth chronologies’ within the hard, calcified tissues of aquatic species, such as fish otoliths, can provide valuable information about how an individual grows throughout its lifetime and what key environmental variables influence that growth. We examine and compare broad and local scale drivers of the growth of black bream (*Acanthopagrus butcheri*), an economically important fish species found throughout temperate estuaries in southern Australia, using otolith growth chronologies. Sectioned otoliths were sourced from two pre-existing collections representing two climatically divergent regions in south-east

Australia: the Lower Lakes-Coorong estuarine system in South Australia, characterised by a relatively warm, dry climate, and the east coast estuaries of Tasmania, characterised by a relatively cool, wet climate. Annual otolith growth increments were counted and measured in fish from 7 to 23 years old, encompassing the period from 1985 to 2010. Using a mixed modelling approach, we relate inter-annual growth variation to local scale changes in air temperature (proxy for water temperature) and rainfall (proxy for freshwater flows) and broad scale changes in relevant climatic indices (Multivariate ENSO Index and Southern Oscillation Index). Based on this analysis, we make predictions on how the growth of black bream, and thus the productivity of bream populations, may be impacted in the future based on climate change projections. This study further validates the use of growth chronologies, a new and emerging field of science, as a valuable tool for understanding long-term ecological responses to environmental change in aquatic systems.

Abstract reference: IC_Kelly_09

Comparison of Slimy Sculpin (*Cottus cognatus*) annual growth in contrasting regulated and unregulated riverine environments

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Hydroelectric dams alter downstream physical conditions, such as water discharge and temperature, imposing alterations that can impact resident biological communities, including fish populations. Slimy Sculpin (*Cottus cognatus*) is a small bodied benthic freshwater fish abundant throughout Canada, with a known limited home range size that facilitates determination of the biological impacts of environmental disturbances at relatively fine spatial scales. Here Slimy Sculpin growth is used to assess the effects of two different riverine flow environments within a common Boreal eco-region of northern Canada. One flow regime is dominated by a 15MW hydroelectric dam where the effects of variation in the dam operational regime (restricted versus unrestricted rates of discharge increase and decrease) downstream of the dam are assessed. The second flow regime is that of a comparably sized natural river. Annual Slimy Sculpin growth was determined by way of otolith back calculations using the scale proportional hypothesis. Slimy Sculpin growth was higher, but more variable on the regulated river relative to the unregulated river and there was no significant mean effect of dam operating regime on growth in the regulated river. There was a significant relationship between sculpin growth and distance from the dam on the regulated river, with growth decreasing with increasing distance. Both sampling site and growth year had a significant effect on first year growth for fish living in both rivers and second year growth for fish living in the unregulated river. A significant effect of sampling site on second year growth in the regulated river suggested greater heterogeneity in growth conditions as a result of flow regulation. Discharge parameters related to volume and frequency tended to be positively correlated with growth in the regulated river, but uncorrelated to growth in the unregulated river.

OtoChuki Session (ID)

Abstract reference: ID_Neville_01

Migrating Atlantic cod otoliths reflect movement through water masses: linking data storage tag data with high resolution trace element and isotope geochemical signatures

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Geochemical signatures are frequently used in otolith research to reconstruct the ambient conditions experienced by a fish. Oscillations in $\delta^{18}\text{O}$ and Sr/Ca corresponding to alternating opaque and translucent aragonite growth bands have been reported in several marine fishes. Both $\delta^{18}\text{O}$ and Sr/Ca in otolith aragonite have been reported to be inversely correlated with temperature, however, other factors (kinetic, physiological or ontogenetic effects) may influence this correlation. In this study we compared the geochemical patterns of fish moving (migrating) through water masses, with those experienced by sedentary (penned) fish. The objective of this was to investigate typical seasonal oscillations in trace element and isotopic signatures for non-migrating fish to elucidate patterns that indicate true fish movements. To do this, we linked ambient temperature from a migrating Atlantic cod from a recaptured data storage tag, (DST) with otolith $\delta^{18}\text{O}$ and trace element (Sr/Ca, Mg/Ca, Ba/Ca, Na/Ca) analyses. The same analyses were run on a number of penned cod with known ambient conditions. DST records coupled with the high spatial resolution analyses permitted temperature calibrations for both Sr/Ca and $\delta^{18}\text{O}$. An increase in the amplitude of annual Sr/Ca maxima and minima occurred at approximately age 3 in the wild cod, consistent with the onset of inshore-offshore migratory behaviour. A change in the amplitude of Sr/Ca was not observed in the penned cod. These findings give unique insight into geochemical fluctuations observed in migrating fish as distinct from the oscillations experienced by resident or penned fish.

Abstract reference: ID_Matteo_03

Settlement synchronization in the red mullet (*Mullus barbatus*) as inferred from otolith sclerochronology

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Exploited fish populations are often limited by recruitment. Settlement is a critical phase in the life history of benthic fishes. Therefore, the settlement process is likely to control the dynamics of recruitment-limited populations to a large extent. This is specially the case in heavily exploited demersal populations of marine fishes. Despite the central importance of recruitment in population dynamics and fisheries science, the available information about settlement of commercially exploited species is scanty. Here, settlement of the striped mullet (*Mullus barbatus*), one of the

most heavily fished species in the Mediterranean Sea, was characterized through sclerochronology of otolith from post-larval specimens.

Spawning date (SPD) and pelagic larval duration (PLD) were inferred from otolith readings.

Temporal variability among and within the 3 studied areas was then determined. Size at settlement (SAS) was estimated from the radius between the core and the settlement mark. SAS was then used to assess relationships between early life-history traits and larval growth.

The present findings showed that the Mediterranean population of *Mullus barbatus* displays wide variability in the duration of the pelagic larval stage. Estimations of early life history traits confirmed such variability, supporting the hypothesis that red mullets start their larval phase in different pulses within a wide temporal window, which results larger than previously thought.

Abstract reference: ID_Mazloumi_05

Growth of King George whiting: an investigation of climatic influences on otolith growth

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Otoliths of fish can provide long-term chronologies of growth, but few such chronologies have been developed for fish in the Southern Hemisphere. Differences in the width of the growth increments of otoliths reflect both growth and environmental variability over the lifespan of the fish. Using King George whiting, a commercially and recreationally important fish species, collected from Kangaroo Island, Gulf St Vincent and Spencer Gulf South Australia, we developed an otolith chronology derived from growth increment widths. Our growth chronology showed synchronous patterns of growth among fish and spanned 22 years (1980-2012). We then compared the otolith chronology to some factors included (Region, Recruitment, sea surface temperature (SST) and Southern Oscillation Index (SOI)) using 'Mixed effects Model'. Similar relations are found for otolith chronologies of King George whiting collected from other regions. No significant effects were found between the otolith increment and other environmental factors. An understanding of the link between climate and growth of fish in temperate regions may be used to predict future patterns in productivity and status of fish stocks under a changing climate.

Abstract reference: ID_Norcross_06

Otolith chemistry discriminates water mass occupancy of Arctic fishes in the Chukchi Sea

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Microchemistry of otoliths can be used to reconstruct fish movement patterns and habitat use. We applied this tool to three abundant western Arctic marine fishes, Arctic/Polar Cod (*Boreogadus saida*), Arctic Staghorn Sculpin (*Gymnocanthus tricuspis*), and Bering Flounder (*Hippoglossoides robustus*). In summer 2009 and 2010 these fishes inhabited three bottom water masses in the Chukchi Sea: Alaska Coastal Water (ACW), Bering Sea Water (BSW), and Winter Water (WW). We tested the relationship between water mass from which a fish was collected and microchemistry of the most recent growth edge of the fish's otolith using Mg, Sr, Ba and Ca. Though not all water

masses could be differentiated by all elements in all fish species, for every fish species at least one difference could be detected. The Mg:Ca ratio was different between ACW and BSW for all three fish species, and between each of those water masses and WW for *B. saida*. Ba:Ca and Sr:Ca could differentiate WW from ACW and BSW in *B. saida*, and Sr:Ca separated ACW and BSW only in *H. robustus*. Temperature and fish age, but not salinity or barium in bottom water, explained differences in otolith chemistry between bottom water masses. Significant variables, trends, and water mass differences were fairly consistent among fish species. A discriminant function post-hoc analysis of fish occupying bottom water masses resulted in 76% correct classification of *B. saida* into bottom water masses when ages were pooled, and as high as 87% classification by separating age classes. Similar classifications were found for the water masses that could be discriminated in otoliths of *G. tricuspis* and *H. robustus*. Use of otolith microchemistry to determine occupancy of water masses over time is most promising for *B. saida* which is widespread in the Chukchi Sea.

Abstract reference: ID_Black_07

Otolith biochronologies and the present, past, and future of the California Current upwelling system

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Biochronologies derived from otolith growth-increment widths are annually resolved, multi-decadal in length, and contribute to the development of multivariate ecosystem indicators. In the central California Current upwelling zone (CC) along the west coast of North America, biochronologies of splitnose rockfish (*Sebastes diploproa*) yelloweye rockfish (*S. ruberrimus*), and Chinook salmon (*Oncorhynchus tshawytscha*) are integrated with indices of reproductive success for common murre (*Uria aalge*) and Cassin's auklet (*Ptychoramphus aleuticus*). The leading principal component (1978-2001) explains 59% of the variability in these time series and correlates strongly and positively to winter upwelling. The second principal component captures an additional 16% of variance and positively relates to summer upwelling. These biological patterns reveal two seasonal upwelling "modes": a summer mode dominated by low-frequency processes including long-term increases within the central CC and a winter mode dominated by higher-frequency, interannual variability. The winter mode is linked to the intensity of the winter North Pacific High (NPH); clockwise circulation promotes upwelling-favorable winds. The NPH also blocks or facilitates the onshore flow of precipitation with impacts on terrestrial productivity. Thus, tree growth is highly sensitive to atmospheric drivers of upwelling and can be used to hind-cast this biologically relevant climate variable over the past 600 years. These same winter atmospheric variables can also be targeted in atmosphere-ocean coupled global circulation models to estimate future variability. In so doing, otolith biochronologies in combination with other proxies can help identify key climate drivers of ecosystem productivity, benchmark their pre-industrial ranges of variability, and identify biologically important variables for forecasting under various emissions scenarios.

Abstract reference: ID_Robillard_08

Life-history connectivity and climate-related change in distribution of red hake (*Urophycis chuss*) on the northeast United States continental shelf

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We use a multidisciplinary approach consisting of Lagrangian particle simulations, otolith chemistry, and age structure analysis, to address population-level effects of climate-related change in red hake (*Urophycis chuss*) and test a management hypothesis of stock separation between the Gulf of Maine (GOM) and Mid-Atlantic Bight (MAB). Lagrangian particle simulations predicted regional retention of pelagic early stages and age structure showed strong spatial heterogeneity, consistent with stock separation. However, the chemistry of the otolith nuclei showed no differences despite sharp environmental gradients. These results can be explained by recruitment from a common source during early life with subsequent separation into groups with divergent mortality. Evidence of similar regional growth rates and environmental exposures from age-at-length and edge chemistry corroborated adult restriction to temperature-favorable habitat, accounting for higher mortality in the more rapidly changing environment of the MAB. We hypothesize that the MAB stock depends on spawning from the Georges Bank: current distributions of red hake in the northern extent of the United States shelf are likely to show stability, at least in the near term, because the critical life history events occupy an area that is removed from the temperature limitations of the species and, relative to the MAB, shows less marked change linked to climate.

Abstract reference: ID_Vanhove_09

Temperature and oxygen isotope variability of North Sea Basin shelf waters during the early Eocene, recorded by fish otoliths

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Shallow marine settings comprise invaluable information about the impact of paleoenvironmental change on shelf ecosystems. We measured $\delta^{18}\text{O}$ and $\delta^{13}\text{C}$ ratios in fossil fish otoliths to assess temperature and salinity variability in the Ypresian (Ypr., 56.0-47.8 Ma) southern North Sea Basin (sNSB). Well-preserved otoliths were selected representing different environmental facies: the marginal marine Blackheath and Harwich Formations (lower Ypr.), the outer neritic London and Kortrijk Formations (middle Ypr.) and inner neritic Tielt Formation (middle to upper Ypr.). Otoliths from demersal and presumably non-migratory fish such as ophidiids, bythitids and congrid were

used. Bottom water paleotemperatures are determined from outer neritic samples, based on bulk sampling. This facies is composed of fine silts and clays deposited in an offshore environment, unlikely to be influenced by freshwater influx. Mean bulk $\delta^{18}\text{O}$ values reveal within-horizon consistency among different co-occurring taxa. The horizon means range between -1.9 and -3.4 ‰. This translates to warm, albeit not extreme MAT's between 21.3 and 28.5 °C during the 54-51.5 Ma interval. Based on incremental sampling, low seasonality of 3-6 °C is observed in the outer neritic facies. By contrast, mean bulk $\delta^{18}\text{O}$ values from the Harwich and Blackheath Fms. are very negative, ranging from -5 to -7 ‰, indicating substantial mixing with low-salinity waters reduced by 3-4 ‰. Restricted connection of the sNSB with the North Atlantic and an intense global water cycle shortly after the PETM may explain this probably basin-wide freshening. Samples from the inner neritic Tilt Fm. bear values between -3 and -5 ‰ and are thought to be partly influenced by freshwater runoff. All inner neritic facies are characterized by a high intra-annual variability of 3-4 ‰, probably the result of seasonally paced freshwater influx. In summary, otolith stable isotope ratios seem to reliably record paleoenvironmental conditions.

Abstract reference: ID_Bareille_10

Does Ba:Ca and Sr:Ca ratios always reflect ambient water composition where Atlantic salmon (*Salmo salar*) inhabits?

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Otolith chemistry has emerged as a useful tool for determining migration patterns, nursery habitats and natal origins of anadromous and freshwater fishes. The accuracy of this tool largely depends on the implicit assumption that the chemical signature of the otolith reflects the geochemical signature of the water in which the fish resides. Numerous studies have established direct correlation between environmental exposure and otolith chemistry especially for Sr:Ca and Ba:Ca ratios but only for a short period of time in the life of fish. However, ontogenetic changes in physiology, environmental variations, or movements may have been confounded as elemental incorporation dynamics and may vary depending on the life history stage of the fish. Thus, endogenous control must be taken into account when interpreting life history transects across otoliths. The current study combined otolith chemistry profiles from known life-history juveniles of Atlantic salmon (*Salmo salar*) (hatchery-reared, field-collected) and geochemical water database to investigate the extent to which Ba:Ca and Sr:Ca variations are associated to the ambient water chemistry or not. This study aimed at improving the interpretation of field-based studies during freshwater residency. The results revealed that Ba:Ca ratio in otoliths was not always determined by its respective metal:Ca ratio in the ambient freshwater. Indeed, Ba:Ca otolith profiles were characterized by some peaks which could not be explained by chemical changes in the ambient water environment. These peak patterns were first observed at the yolk sac absorption mark but also when fish was moved from a hatchery to a river as well as following the smoltification event. At the yolk sac absorption mark, both wild-caught and hatchery-reared juveniles exhibited a Ba:Ca peak pattern, followed by a long tail-off until ratio remained stable and related to ambient water. We suggested that physiological processes are largely involved in the complex response of otolith Ba:Ca ratio.

Comments on the microstructure of vertebrae, and estimates of age and growth of the blue shark, *Prionace glauca* (L., 1758), in the southwest Atlantic

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The age structure of *Prionace glauca* in the southwest Atlantic was estimated from vertebrae collected aboard two commercial longline fishing vessels in southern Brazil and adjacent international waters (c.a. 23°-38°S; 29°-52°W). Fork length (LF) was measured in 2340 males (84-262cm) and 402 females (73-247cm). Vertebral frontal plane sections for light microscopy (LM) (7-21µm) and stereoscopic microscopy (SM) (0.75mm) were stained (Alizarin, Alcian-Blue, Hematoxylin and Silver Nitrate) for permanent mounting in Canada balsam. Only alizarin and silver nitrate improve pair-band recognition, allowing us to define the growth marks (GM) as the only point at which the pattern of calcification switches, from more calcified to less calcified. In LM, the GM were discernible from 21µm sections. Hyaline cartilage was found adjacent to perichondrium and in two spheroid bodies near to the focus. This last hyaline tissue seems to contribute to the interstitial growth from isogenic groups. However, interpretations have been based in the cartilage tissue of perichondrium. Therefore, further study is necessary. In the SM vertebral sections of 260 females and 677 males were analyzed. Vertebral sections were read twice. Ageing precision was analyzed through bias graphs and precision indexes both, for the entire sample and by age groups (D=8.07, APE=11.42). Marginal increment analysis revealed that GM are formed annually in winter. The maximum ages were 12.5 and 13.6 years for females and males, respectively. VBGF was adequate to describe growth, with no differences between sexes ($L_{\infty}=265,2$, $k=0,149$, $t_0=-1,504$). In the first year of life *P. glauca* doubles in size, from 45cm to approximately 90cm. The age structure of catches consists primarily of 4-5 yr-old subadult males, and 6 yr-old adult females. Small juveniles of 1 to 3 years old occur south of the Subtropical Convergence, whereas large adults, with more than 10 years, are scarce throughout the area.

Poster Session (IE)

Abstract reference: IE_Labonne_01

Characterization of otolith microchemical signatures from 3 fish species along the Moroccan coast: link between anthropogenic vs. natural influences and trophic level

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The Canary Current Large Ecosystem is characterized by a major, nutrient-rich up-welling of deep, cold oceanic waters, which stimulates high biological productivity that results in an abundance of both pelagic and demersal fishery resources. In this area, the Moroccan coast is also under direct anthropogenic activities influences such as phosphates industries. Several observations indicate that variations of the upwelling activities observed in the recent years would generate the remobilization of the contaminants. Our study deals with the metallic concentrations in the otoliths of 3 fish species of economic interest (*Merluccius merluccius*, *Pagellus acarne*, *Sardina pilchardus*) along the Moroccan coast. The fishes were collected in markets in June 2012 for the northern part of Morocco (during the maximum of the upwelling activity) and in November 2012 for the southern part. Concentrations were measured by ICP-MS on the whole otoliths to characterize environmental signatures. Samples from a campagne at sea in June 2013 were also analyzed. First results indicate that the concentrations are different from one species to another and are slightly different from north to south. Concentrations in Barium and Uranium, are higher in samples from the south and Cadmium is only detectable in *S.pilchardus* from this area. Aeolian Saharan dust signature is also visible in these samples. Stock discrimination will be also discussed based on otolith chemistry.

Abstract reference: IE_Janowska_02

Chemical composition of fish otoliths from a lake subject to reclamation

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The subject of the study were planktivorous and prey fish originating in Trzesiecko Lake, which due to its poor ecological state and emerging blooms of cyanobacteria, underwent efforts at reclamation. Several methods were deployed: application of ferric sulphate, aeration of the deep water and biomanipulation consisting of exerting pressure using predators, among others perch

that would prey on plankton feeding fish. Chemical composition analysis of the otoliths was performed by means of a scanning electron microscope by JEOL with an EDS analyser. Each of the otoliths was rinsed with deionised water prior to the analysis. The analysis was conducted in one plane from the right side of the otolith to the left one. Three places were selected for detailed chemical composition analysis. They showed very high percent weight and percent mass contribution of the following elements: carbon, calcium, oxygen, and nitrogen. Varied amounts of other elements were also recorded, including metals such as: aluminium, manganese, chromium, molybdenum, or iron. The presence of these metals was confirmed via graphite furnace atomic absorption spectrometry (GFAAS) of the analyzed otoliths. Under chemical analysis of the otoliths it was especially disturbing to confirm the presence of iron atoms which may suggest that iron deriving from iron sulphate application may overcome biological barriers and accumulate in the organisms causing disturbances in their metabolism.

Abstract reference: IE_Reis-Santos_03

Effects of temperature and water composition on otolith chemistry across a salinity gradient

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Elemental composition of fish otoliths provides a valuable means to retrospectively determine a fish's environmental history over time. However, to reconstruct the patterns of fish movement and migration using otolith chemistry it is essential to establish the link between changing environmental variables, in particular, temperature, salinity and elemental concentration, and the concentration of elements within otoliths. Using a controlled laboratory experiment we investigated how otolith chemical composition (Mg:Ca, Mn:Ca, Sr:Ca, Ba:Ca) was simultaneously affected by temperature, salinity and ambient elemental concentration in juvenile European sea bass *Dicentrarchus labrax*. A total of 18 treatments representing all combinations of temperature (21, 25 °C), salinity (10, 20, 30) and ambient water concentrations (Low, Medium, High) were analysed. Otolith elemental composition of Sr:Ca and Ba:Ca were positively related to ambient water concentration. Consistent interactions between salinity and concentration were found for otolith composition, as well as elemental incorporation. Positive effects of temperature on elemental composition and incorporation were also detected. The partition coefficient of Ba (DBa) initially decreased with increasing water concentration but increased at the highest level of added elemental concentration. Across salinity gradients, DBa increased whilst distinct patterns of variation occurred for the partition factor of remaining elements within water concentration treatments. The importance of validation experiments to decipher species-specific patterns in otolith chemistry to describe fish movement and habitat use patterns are discussed.

Abstract reference: IE_Limburg_04

Fish Tales Through Fish Ears

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Otoliths are valuable for scientific research. Yet they also possess a kind of beauty. Visualization of otolith chemistry via 2-D trace elemental mapping can also produce interesting and appealing images. This project, intended as an art book, will extend my science as art for a broad audience, whether scientists or not. Through focusing on a number of fish species from different parts of the world, with different life histories and ecology, I will let otoliths "speak" to reveal the "tales" of fishes.

Abstract reference: IE_Agiadi_05

How did past environmental change control the distribution of small pelagic fish in the Mediterranean Sea? Examples from the fossil record

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Environmental variability determines fish distribution, migration and abundance both in the present as well as in the past. The close link between climate and fish populations has been observed in modern and historic times. The past distribution of small pelagic fish species, such as sardines and anchovies, is especially of interest since they have repeatedly been proven to respond rapidly to climatic variability. These fish generally have short life spans; their abundances are strongly driven by the annual recruitment of young fish, a process modulated by ocean climate. New data on Mediterranean fish paleobiogeography shows that naturally-occurring changes in the geological past have repeatedly modified the fish distribution in this area. Here, the geographic distributions of *Engraulis encrasicolus* and *Sardina pilchardus* are compiled and examined with regard to the global, regional and local paleoenvironmental conditions in order to draw conclusions as to the parameters affecting them. Anchovy remains have been found in coastal deposits from the Messinian (Kalamaki section, Zakynthos Island, Ionian Sea), the Gelasian and the Calabrian (Ypsenis section, Rhodes Island, southeastern Aegean Sea) and the Ionian stage (Fiumefreddo section, Italy; Kallithea section, Rhodes Island, southeastern Aegean Sea), where the presence of this species is often associated with a climatic optimum conditions. *Sardina pilchardus* is also known from the Messinian stage (Tanaro river, northern Italy), the Zanclean stage (Agia Triada section, Peloponnese, southern Greece) and the Calabrian stage (Gravina section, southern Italy). The fossil otolith record of anchovies and sardines in the Mediterranean realm reveals a consistent pattern of migrations and re-establishments of their populations from the Miocene until today. This research has been co-financed by the European Union (European Social Fund – ESF) and Greek national funds through the Operational Program "Education and Lifelong Learning" of the National Strategic Reference Framework (NSRF) - Research Funding Program: THALIS – UOA – MEDSALC.

Abstract reference: IE_Songer_06

Investigating the distribution of crystalline otoliths

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An investigation of the spatial distribution of crystalline otoliths in UK waters, using material from Cefas otolith archive. Initial thoughts on possible causes for this distribution.

Abstract reference: IE_Górski_07

Migratory life-history patterns of *Galaxias maculatus* in the Southern Hemisphere rivers revealed by otolith microchemistry

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The analyses of microchemical composition of otoliths was used to establish the movements of *Galaxias maculatus* between freshwater and marine/estuarine environments in two river systems at similar latitude – the Waikato River in New Zealand and Valdivia River in Chile. Fish and water samples were collected from various habitats within both river systems (the main channel, tributaries and riverine lakes). Otoliths and water samples were analysed for a suite of trace elements using Laser Ablation Inductively Coupled Plasma Mass Spectrometry. Otolith elemental composition was analysed along a transect from the core to the edge of each otolith. We found significant differences in migratory life history patterns between *G. maculatus* from the Valdivia River and the Waikato River. *Galaxias maculatus* from the Waikato River appeared to have recruited mostly in the ocean as indicated by high strontium (Sr) to calcium (Ca) ratios in the core region of the otoliths of analysed fish. Conversely, the majority of *G. maculatus* from the Valdivia River recruited in freshwater habitats. Furthermore, in the Valdivia River also fish collected in the estuary displayed low Sr/Ca ratios in the core region of the otoliths suggesting upstream migrations to freshwater habitats for spawning. We hypothesise that these differences in migratory life histories between the two rivers are an effect of different availability of nursery habitats with more lentic and wetland habitats in the Valdivia River. (Funding: Fondecyt 3130690)

Abstract reference: IE_Helser_08

Modelling Environmental Factors Affecting Assimilation of Bomb-produced $\Delta^{14}\text{C}$ in the North Pacific Ocean: Implications for age validation studies

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The bomb-produced radiocarbon (^{14}C) chronometer has become the gold standard for assessing the accuracy of otolith growth ring based fish age estimates. In the northeast Pacific Ocean, nearly a dozen age validation studies have been conducted, ranging from California to Alaska, most of which have relied on a single reference chronology from the Gulf of Alaska. We developed a Bayesian hierarchical model using data sets of bomb-produced radiocarbon in the northeast Pacific Ocean and investigated whether latitude and upwelling exerts an influence on the parameters that describe the rapid $\Delta^{14}\text{C}$ increase in marine calcium carbonates. Models incorporating both latitude and upwelling as linear covariates of a 4-parameter logistic model were favored based on ΔDIC statistics. There was substantial evidence to support that the timing of the $\Delta^{14}\text{C}$ pulse was advanced and that total $\Delta^{14}\text{C}$ uptake increased with increasing latitude. In contrast, increased oceanographic upwelling resulted in lower total radiocarbon input as well as a delay in the timing of the pulse curve, as was demonstrated in the upwelling dominated California Current System. Within the observed latitudinal and upwelling range of the data sets examined in this study the predicted timing of the bomb pulse curve varied by as much as 3 years, which could be misinterpreted as ageing error. Our results suggest that new reference chronologies may be needed for regions of the North Pacific Ocean differing in latitude, seasonal upwelling strength and other mixing factors that can potentially change the functional form of the $\Delta^{14}\text{C}$ curve.

Abstract reference: IE_Paulus_09

Namibian hake *M. capensis* otolith zonation related to fish and environmental conditions in the northern Benguela

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In this study, we describe the periodicity of translucent zone formation and analyse the link between zone formation and environmental and biological variables of one of the Namibian hake species, the shallow-water hake *M. capensis* in the northern Benguela upwelling ecosystem. Otoliths of 1300 *M. capensis* were collected from the fishery to represent each month of the year

2007–2013. For each sample, each translucent zone present was measured irrespective of indicating an annulus. From marginal increment analysis, we show that 2–3 translucent zones are formed annually, 1–2 between austral summer and autumn (peaks in February and May) and one in winter–spring (peaks in August–September). We further investigate the ecology of these fish in relation to otolith zonation. Translucent zone formation “events” were back-calculated and linked to the environmental variables (bottom and sea surface temperature, dissolved oxygen and salinity) and biological variables (fish condition, gonad stage / GSI, and liver weight) at the time of zone formation using generalised linear modelling. No significant temporal correlation with any of the effects was found, but rather a combination of each effect played a role. In the cold temperate system of the northern Benguela, otolith zonation is not as predictable as generally assumed. Zones were associated with high GSI or high liver weight, poor fish condition and warm temperatures or low GSI and good fish condition at cold temperatures. Liver weight and salinity also seem to play a role in the formation of second to the fifth translucent zones. This is evidence that zonation, one of the main applications of otolith research is still poorly understood and more emphasis should be placed on this in all regions.

Abstract reference: IE_McFadden_10

Otolith Biomineralisation: Insights From a Microstructural and Microanalytical Study

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Elements such as Sr which are thought to be environmentally regulated, are of particular interest in the reconstruction of environmental conditions such as temperature histories. Recent studies of the shells of the bivalve *Arctica Islandica* however revealed an association between the Sr/Ca, Ba/Ca and Mg/Ca ratios and crystal fabric. In particular, the heterogenous variation of Sr in the shells did not reflect environmental changes but rather showed an association to the crystal size and shape. Changes in Sr/Ca ratio, unrelated to environmental conditions, have been observed following a metamorphosis check in eel otoliths, similar to that observed in the annual growth checks in bivalve shells. These observations support the existence of a relationship between trace element concentrations (in particular Sr concentrations), and the microstructure of the otolith. In conjunction with microstructural interpretation, variation of the Sr/Ca ratio in otoliths of *Platycephalus bassensis* have been determined in an effort to uncover the role of biomineralisation in otolith trace element uptake. Microstructural analysis was undertaken by Scanning Electron Microscopy (SEM) and Electron Backscatter Diffraction (EBSD), and subsequently Sr/Ca ratios across otolith growth increments were determined by Electron Probe Microanalysis (EPMA). Sr/Ca values were heterogeneously distributed across the otolith growth increments. SEM images and EBSD orientation data reveal the changes in otolith grain structure appear to be associated with the changing Sr/Ca values. The correlation between Sr/Ca ratio and grain size was indicative that Sr is not completely regulated by the environment, but is instead controlled by a complex interaction of biological and kinetic effects.

Abstract reference: IE_Arechavala-Lopez_11

Otoliths as indicators of wild or farm origin of sea bream and sea bass

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Farming sea bream (*Sparus aurata* L.) and sea bass (*Dicentrarchus labrax* L.) in open-sea cages has risen during the last decades, and consequently, problems also began to arise concerning escape events. Escapees might lead to economic losses and environmental problems in coastal ecosystems, which might be monitored and evaluated to further management strategies. Thus, it is necessary to develop indicators to distinguish between the wild and farm origin of such escapees within native populations. Based on the fact that cultured fish grow in a different environment from in the wild, feeding on different trophic resources and resulting in faster growth and frequently different growth patterns, it might be expected that they also show morphological and chemical differences according to fish origin in sagittal otoliths. To test this, sea bream and sea bass individuals from farm facilities and wild assemblages were sampled in Spain and Greece. Variation in otolith morphology was examined through conventional shape descriptors (*i.e.* area, perimeter, circularity, roundness, mass, and height-length relationship) and elliptic Fourier descriptors (EFDs). Discriminant analysis with either all shape descriptors together or EFDs was able to classify with high accuracy both sea bream (89.5-95.7%) and sea bass (93.2-95.2%) according to their origin. Variations in trace element compositions (*i.e.* Mg, Al, K, Mn, Fe, Co, Ni, Cu, Zn, Sr, Ag, Ba, Pb) between fish origin for both species were also analysed. Classification through discriminant analysis with 13 trace elements showed high percentages regarding the wild and farm origin, for both sea bream (83.3-89.8%) and sea bass (87-100%). Hence, this study recommends the use of morphological characteristics and trace elements compositions of otoliths as useful indicators of sea bream and sea bass origin, and therefore, as fisheries management tools.

Abstract reference: IE_Limburg_12

Paleo- and modern salinity and temperatures as recorded by Baltic Sea cod *Gadus morhua* over millennia

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The Baltic Sea has experienced variations in its salinity and temperatures over its ca. 8000 year existence, due to climatic factors such as warming and cooling periods as well as to intrusions of North Sea water or retreats due to post-glacial isostatic rebound. We examined cod otoliths and vertebral bones from five periods: two periods in the Neolithic Age (4400–3800 y.b.p.), Iron Age (1000 y.b.p.), latter 1600s (Sweden's Empire Period), and modern collections from periods with low (1990s) and high (2000s) hypoxia. ^{87/86}Sr was quantified to fix salinities, and then $\delta^{18}\text{O}$ was used to determine temperatures during those time periods.

Abstract reference: IE_Fabré_13

Pattern of otolith growth in earlier stages of *Mugil liza* in tropical and temperate regions of Southwestern Atlantic Ocean

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Fish somatic growth is reflected in otolith growth and check-marks pattern, which are influenced by environmental factors. Increase in otolith size of pre-adult depends on the birth date and growth rate, which are influenced by the environmental factors, such as temperature, salinity and food availability among others, in the nurseries and breeding areas. One of the most important species in the southwestern Atlantic Ocean coast is *Mugil liza*. The purpose of this study was to analyze marks pattern in earlier stages of *M. liza* in a tropical region (Mundau Lagoon, 09° 37' 59,8" S 035° 46,10" W, Brazil) and a temperate region (Bahía San Blas, 40° 33'S- 62 °11'W, Argentina). Radial distances of pre-annual rings (c.a. rings present before the first sexual maturity) of fish otoliths from both regions (Brazil, N = 380; and Argentina N = 210) were measured. Significant differences between both regions were determined for each of the three pre-annual otoliths rings. These differences could be associated to environmental dissimilarities in both regions during the life the specimens considered.

Abstract reference: IE_Evans_14

Tracking Stocked European Eel (*Anguilla anguilla*) using Otolith Microchemistry

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Recruitment levels in the European eel, *Anguilla anguilla* (L.) have declined to <5% compared to pre-1980 levels with numerous causative hypotheses including exploitation, habitat destruction, pollution and parasite introduction. The eel is now listed as critically endangered by the International Union for Conservation of Nature (IUCN), prompting EU action taking the form of the EU Eel Recovery Plan (1100/2007). Member states had to prepare an Eel Management Plan (EMP) aimed at bringing about stock recovery via reducing anthropogenic impacts and increasing spawners migrating to sea. One option available to EMP's was the stocking of juvenile eel into habitats devoid of migration barriers. Stocking eels from a small number of donor estuaries concentrated around the Bay of Biscay and SW England to EU Nations is considered to be controversial, lacking sufficient evidence of success yet high in risk in terms of pathogen transmission and potential loss of DNA integrity. Since 2009 many EU nations have advocated the

use of stocking as a tool in their EMP's, prompting calls from the EU Commission for robust evidence as to the value of stocking. As such the development of a rapid tracking system was required which was not dependent upon examining Sr marked otoliths over the life of an eel up to 20 years later post-marking. Trace element incorporation into fish otoliths varies among samples collected from different locations. If otolith elemental composition ("fingerprint") somehow reflects the characteristic of the ambient water the fingerprint of the otolith nucleus could serve as a natural marker of eel derived from different donor estuaries. Laser ablation-inductively coupled plasma mass spectroscopy (LA-ICPMS) was used to assay the concentrations of 14 isotopes in glass eel otolith nuclei from different regions of the EU. Results, their implications for Eel management and application towards assessing the value of stocking are discussed.

Abstract reference: IE_Volpedo_15

Use of lapillus otolith microchemistry as indicator of the habitat of *Genidens barbatus* in different estuarine environments from the southwestern Atlantic Ocean

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The marine catfish *Genidens barbatus* is an anadromous species found in South America. The objectives of the present study were to evaluate a new method consisting in the partial polishing and analysis of the microchemistry of the lapilli as indicator of the habitat of *G. barbatus* and identify the potential breeding areas and fish stocks between the populations from the lower section of the Plata Basin (De la Plata River estuary-Argentina) and Lagoa dos Patos coastal lagoon (southern Brazil). Sr:Ca, Ba:Ca and Mg:Ca ratios were measured in the core area (inner 4 rings) and external area (outer 3-4 rings) of the otoliths by ICP-OES. The Sr:Ca ratio tended to be higher in the otolith external area than in the core area, while the Ba:Ca ratio followed the opposite pattern, suggesting the moving towards areas of higher salinity. The results show significant differences ($p < 0.05$) in the Sr:Ca, Ba:Ca, Mg:Ca ratios in the core and external area of otoliths from the Plata Basin and those from Lagoa dos Patos that may indicate the occurrence of at least two different breeding sites and suggests the occurrence of at least two fish stocks in the region.

Abstract reference: IE_Heimbrand_16

Using otoliths to distinguish sea spawning from river spawning whitefish (*Coregonus maraena*)

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In the Baltic Sea two major reproductive forms of whitefish (*Coregonus maraena*) are recognized; a migratory form spawning in coastal rivers and a more resident form spawning in shallow bays. Although these ecological forms differ somewhat morphologically there is a large overlap in

characters and no clear-cut way to differentiate between them. The drastic decline of whitefish in the northern Baltic Sea in recent years highlights the need to differentiate between spawning types in order to make reliable assessment and take effective management actions. By using the chemical composition of otoliths as indicator of the environment the life history of a fish can be revealed. Otoliths are skeletal structures of the inner ear that grow during the life of the fish while incorporating elements from the surrounding water. Since the ratio of Strontium and Calcium are related to the salinity of the water the Sr/Ca ratio in the otolith could be used to distinguish between marine and freshwater origin of the fish. In an experiment, otoliths were prepared from 7 adult whitefish from the river Gideälven, and 12 adults from the Söderhamn archipelago. PIXE analysis of polished otoliths mounted on an object glass revealed that despite sampling during spawning time in supposedly different spawning habitats there was a mix of fish that originated from river spawners and sea spawners in both areas. Only 2 of the 7 fish from Gideälven had spent their first year in freshwater while 3 of the 12 fish caught in the archipelago had a freshwater origin.

Abstract reference: IE_Buckthought_17

Utility of otolith chemistry in determining stock structure of grey mullet (*Mugil cephalus*) in northern New Zealand

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Understanding connectivity and stock structure is a fundamental aspect for the management of any fishery. For fish species that inhabit estuarine nursery habitats during the juvenile phase, quantifying the contribution of different nurseries to a fishery is paramount. Otolith chemistry analysis has great potential in this respect as it can often distinguish the natal origin of individual fish. In northern New Zealand, grey mullet (*Mugil cephalus*) are estuarine-dependent during their early juvenile life-stages, potentially producing distinct chemical otolith signatures (tags) that may allow us to better understand stock structure within the grey mullet fishery. We assessed otolith chemistry signatures for juvenile grey mullet from estuaries throughout their mainly northern New Zealand range, using a subset of fish from 9000 individuals collected across 88 estuaries. The main nursery estuaries were assigned to one of seven spatial regions, and quadratic discriminant analyses (QDA) used to develop a tool to allow fish otoliths of unknown origin to be allocated back to one of these regions, based on their chemical signature. The QDA method produced mixed results; high allocation success (up to 72%) was achieved for some regions, but overall success was only 47%. This allows some specific hypotheses to be assessed, but not others. The next step is to collect adult grey mullet in the 2014/15 austral summer, and compare their chemical signatures with known origin juveniles based on some specific testable hypotheses, to better understand stock structure. As adults are wide ranging, occupying estuaries, entirely freshwater rivers/lakes, and exposed coastal regions distant from estuaries; this work will also assess differing life history connectivity strategies.

Where do elements bind within the otoliths of fish?

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Element analyses of otoliths are used extensively to reconstruct environmental histories of fish, based on the assumption that elements substitute for calcium within the CaCO₃ otolith structure. However, evidence suggests that elements may also be incorporated within the organic component of the otolith. Should incorporation within the organic component occur, rather than the direct substitution for calcium, then environmental reconstructions may be misinterpreted. This study determined patterns of element distribution among the organic and inorganic components of otoliths. Organic otolith components (DNA and protein) were isolated and concentrations of a suite of elements were quantified. Elemental concentrations were compared among the organic components, as well as to whole otoliths. Of the elements investigated, those considered 'non-essential' to fish physiology appear to be readily incorporated into the CaCO₃ otolith structure, inferring their suitability for environmental reconstructions. Elements essential to fish physiology were distributed primarily in the organic component of the otolith. These findings enhance our understanding of patterns of element incorporation into otoliths, and ultimately test a fundamental assumption of otolith science. These findings will also improve the accuracy of environmental reconstructions based on otolith chemistry.

Slideshow Poster Session (ISS)

Abstract reference: ISS_Tracey_01

Age and growth study of deepsea coral in aquaria

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As a precursor to conducting laboratory trials on live deepsea corals to investigate growth, resilience, and ocean acidification impacts, a study was carried out to evaluate the feasibility of successfully collecting live specimens at sea and maintaining them in the laboratory. One live colony of the reef-forming scleractinian stony coral (*Solenosmilia variabilis*) was sampled on a voyage to the southern end of Kermadec Arc, and was kept alive at a hatchery facility for 14 months from collection date. The survival period in-aquaria is a world first for this species.

Solenosmilia variabilis appears to be a robust species for in aquaria studies and with several newly sampled coral colonies from the region, further research is now being undertaken to measure growth (radial and linear extension, buoyant weight, and image analysis to observe changes in morphology) to help improve our knowledge of coral growth patterns particularly in understanding the impacts of ocean acidification. We acknowledge that any length increment studies would need to be undertaken for several years as this coral is a very long-lived species. At the end of the experiment, radiocarbon dating will be carried out to determine colony age.

Abstract reference: ISS_Plaza_02

Chronological growth patterns of the endolithic bivalve *Petricola patagonica* associated to a subduction Chilean earthquake

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Coastal uplift produced by large subduction earthquakes cause high mortality of subtidal organisms, which die due to emergence. Hence, the occurrence in supratidal levels of died sessile organisms indicates that a previous tectonic uplift took place. Annual growth bands of these organisms can be cross-matched to the year it was formed to generate time series of growth patterns. A synchrony between calcareous-based growth series and three- ring chronologies are expected due to the coupling between ocean and atmosphere. Consequently, it would be possible to correlate growth chronologies of uplifted specimens with unknown date of occurrence, with large tree-ring chronologies extended into the past. Consequently, the year where both series matches would become a proxy of the year in which the tectonic event with unknown origin occurred. To test this hypothesis, growth chronologies of the endolithic bivalve *Petricola patagonica* were characterized. The specimens were collected in coastal areas uplifted by the giant 1960 Chile earthquake. Internal

growth bands were measured in thin transversal section. An exponential model was fitted for each individual to remove the effect of individual growth. A positive cross-correlation at lag 0 between a growth index (GI) and mean annual sea surface temperature (SST) prior to 1960s demonstrated uplifted specimens of *P. patagonica* were contemporaries, validating that growth bands were formed on an annual basis. Further evidence that collected uplifted specimens died due to the 1690 Chilean earthquake was the existence of significant correlations between GI and a tree-ring chronologies of the Larch *Fitzroya cupressoides*.

Abstract reference: ISS_Hällbom_03

The importance of age determination for assessment of the status in fish fauna

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Fish age is seldom reported in current environmental monitoring programs. We want to show how the age determination of fish in lakes complement the fish indicators included in the current assessment criteria for ecological status. The information is based on fish species frequently occurring in Swedish lakes. We will illustrate this in relation to climate-related factors. Fish growth is often limited by competition both within and between species, which makes it difficult to predict the overall growth changes as the direct effects of environmental changes. Regardless of the indicators used, it is often advantageous to use multiple individual indicators for the interpretation of state and trends. The collection of age data from several lakes with standardized fish sampling increase the possibility of including age based indicators of future assessment. A broader data base can definitely give a more reliable picture of the natural variation.

Abstract reference: ISS_Walker_04

The use of otoliths to compare three species from the Zoarcid family

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Due to climate change and increased industrial development, arctic ecosystems have moved into the spotlight of scientific research. It is important to develop baseline knowledge of the organisms present in this unique environment. During the field seasons of 2011 and 2012, three Zoarcid species were collected using bottom trawl gear. These included Archer Eelpout (*Lycodes sagittarius*), Canadian Eelpout (*Lycodes polaris*), and Longeared Eelpout (*Lycodes seminudus*). These three species were chosen based on their occurrences in bottom trawls and their distribution among depths. Otoliths were removed and prepared for age estimation and trace element analysis. The purpose of this study was to find the differences and similarities between these species and to establish relationships between shelf and slope species in the Beaufort Sea. Age estimations were compared between and within species to determine if there were similarities in age at length between depths and between species. Results suggest that slope species have higher age estimations than shelf species. Otoliths that were not used in age estimation were analyzed for trace elements using laser ablation inductively coupled plasma mass spectrometry (LA-ICPMS).

Comparisons were made between and within species to establish element to calcium ratios and to determine whether trace element signatures were similar among species and sampling locations. Further analysis needs to be done on the trace element transect results to better understand what relationships are present between water depth and otolith chemistry.

Abstract reference: ISS_Wang_05

Tracing the impact of oil spills event by using chemical signatures in squid statoliths and tissues

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The researches to evaluate the impact of ship oil spills events on ecological environment and fishery resources are still limited. In October 2011, M.V. Jui Hsing was stranded at Waimu Shan seashore in northeast Taiwan. It spilled a huge amount of bunker oil that contained heavy metals and polluted the surrounding waters. This study aims to investigate the impact of oil spills event by analyzing the trace elemental concentration in the statoliths and tissues of a dominant squid species, *Uroteuthis (Photoligo) edulis*, collected in nearby waters for finding possible chemical signatures that recorded the anthropogenic event to nature environment. Specimens of *U. (P.) edulis* were collected from June to April in 2011-2012 that covered the period before and after the event. Concentrations of Ba, Ca, Cd, Cu, Fe, K, Mg, Mn, Na, Pb, Sr, and Zn in statoliths, mantle, digestive gland and gill of squids were measured by using ICP-MS. The results showed that the concentrations of Cd in mantle, digestive gland and gill of *U. (P.) edulis* increased after the event. The trace elemental concentration in the statoliths of *U. (P.) edulis* also varied that might be influenced by different water chemistry caused by the events. Chemical signatures of the oil pollution were discovered from statoliths and tissues, however, more research will be needed for quantifying the impacts for more detail evaluation.

Theme II: Community Indicators

Plenary session (IIA)

Abstract reference: IIA_Nolf_Key

Fossil fish otoliths and their importance as indicators of fish communities through time

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One can state that the start of systematical studies on the external morphology of otoliths was established by Koken in 1884, with his study on fossil otoliths from the German Oligocene, which he corroborated with an extensive documentation of comparative Recent material. In fact, all major collections (speaking in terms of systematic diversity) of Recent otoliths were built up by paleontologists, e.g. Weiler (SMF), Stinton (BMNH), Fitch (LACM), Nolf (IRSNB).

The most important data provided by fossil otolith studies certainly are their impact on the fossil record of teleostean fishes: otoliths considerably extend this record in comparison with the classic data provided by osteological remains. PATTERSON (1993) in "The fossil record" provides an overview of all fossil teleost families. At that time, 224 living families were also known by fossils, and 54 of these were exclusively represented by otoliths. Today, the number of such families has increased to 78. Moreover, for many families that have a fossil record for both osteological material and otoliths, the first appearance in time is documented by otoliths. Several taxa have a remarkable otolith-based record, e.g. Gadiforms, Ophidiiforms, Sciaenids, and Gobiids. It is important to consider technical aspects, such as otolith collecting, collection building and otolith storage in order to make best use of these valuable resources.

Abstract reference: IIA_Schulz-Mirbach_01

What do we know about otolith function in hearing? – Insights from cichlids and livebearing fish

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Otolithic endorgans in fish function as accelerometers and serve for detecting both the position and motion of the body and sound. Important factors that influence otolith motion are otolith mass and shape. While it is still widely unknown how different shapes affect otolith movement relative to the sensory epithelium, greater otolith mass was predicted to result in enhanced stimulation of sensory hair cells and improved hearing. Sparse studies on this topic yielded different results in that they did or did not find a tight correlation between heavier otoliths and enhanced hearing. We therefore

investigated the relationship between otolith morphology including (3D) models of otoliths using high-resolution microCT imaging and otolith weight as well as hearing abilities in two model systems of freshwater fishes. We compared i) surface and cave forms of the Atlantic molly (*Poecilia mexicana*) and ii) three cichlid species (*Etroplus maculatus*, *Hemichromis guttatus*, *Steatocranus tinanti*) varying in their swim bladder morphology and connection to the inner ears. Ecotypes of *P. mexicana* had similar hearing abilities and saccular otoliths did not differ significantly in weight. Conversely, the cichlid species showed pronounced differences in hearing abilities with the heaviest saccular otoliths found in the species displaying the best hearing sensitivities. These outcomes seem to support the predicted relationship between otolith weight and hearing abilities. But our findings also highlight the importance of 3D models which are ideally suited for future studies modeling complex otolith motion and thus may provide a better understanding of how otolith morphology contributes to inner ear functions.

Abstract reference: IIA_Shores_02

Stable carbon isotopes in marine fishes otoliths

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Metabolism is a fundamental biological process that controls or explains a wide variety of biological traits and ecological patterns. Metabolic rate (MR) is directly linked to temperature, thus climate change is predicted to impact MR. Due to the inaccessible nature of marine species there are few measurements of field MR available for marine teleost fishes, especially deep-sea species which cannot be maintained in laboratory conditions. The isotopic composition of carbon in otoliths has been linked to MR in several previous studies but to date this link has not been exploited to infer MR in wild fishes. The growth properties of otoliths mean that changes in individual MR through ontogeny could also be determined. Otoliths are widely accessible, and large historic archives exist across global fisheries, potentially allowing tests of changes in field MR in response to temperature or other ecological variables. There is, therefore, a strong incentive to validate otolith carbon isotopes as a proxy for field metabolic rate.

In this study otolith samples from c.150 individuals of fishes inhabiting the UK continental slope between 500 and 2000m were analysed using stable isotope analysis. The relationship between otolith carbon isotope ratios and factors known to influence MR such as body size and temperature was assessed within and between species. Results are promising, suggesting that otolith carbon isotope analysis can be used as a new method of measuring field MR that could be applied retrospectively to teleost fishes.

Abstract reference: IIA_Lombarte_03

Eleven years of the interactive AFORO (Shape analysis of otoliths) database web site (2003/2014)

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In 2003 was created AFORO (<http://aforo.cmima.csic.es>), an interactive system to deal with shape analysis of fish otoliths and a classification system based on the mathematical properties of the one-dimensional curves describing the otolith contours. The system is connected to a database of complete morphometric (measures and morphological indexes) information, including Elliptic Fourier analysis and wavelets (WT) based in otolith contours of the otolith images of well identified samples. Since 2006, AFORO incorporates an expert system that will allow carrying automatic taxon identification based. In 2009 AFORO was included in GBIF (Global Biodiversity Information Facility) databases system, with geographical information of the otolith samples. The AFORO database and public web site (the otolith guide, with its corresponding shape analysis and classification system) keep being continuously implemented. Since 2009, the number of specimens uploaded have increased (from 2492 to 4555 images), and thereby biodiversity representation (from 772 to 1381 species, from 156 to 216 families, from 32 to 36 orders) as well as geographical extension, especially from South Pacific, Central Atlantic and Indic Oceans. Nevertheless, Mediterranean Sea is the best represented area (90% of common species, 64% of species sometimes cited in this area, including 50% of Lessepsian species -Red Sea invaders- are showed). The main advance in the last years was the inclusion of videos of 3D otolith objects in 2012, obtained with LED white light-based scanner, which eliminates the speckle noise effect of laser scanners. This scanner was especially developed for otolith studies. Other novelties are the inclusion of automatic morphometric software and numerical information of Fourier and wavelet analysis, which will be used as morphometric bank for researchers that require precise species identification as paleontologists, archeologists and trophic ecologists.

Abstract reference: IIA_izzo_04

Multi-decadal biochronologies indicate species-specific responses to environmental change

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The long-term influence of environmental fluctuations on estuarine fish populations is often poorly known. Since many species of fish are long-lived, otoliths provide a means of reconstructing past environmental influences on fish populations. We aimed to assess responses in otolith growth to decadal environmental variation in the River Murray estuary, Australia's largest estuarine system, for three fish species with differing ecologies: *Macquaria ambigua* (freshwater obligate), *Acanthopagrus butcheri* (estuarine dependent), and *Argyrosomus japonicus* (estuarine opportunist). Biochronologies based on increment widths in otoliths were developed for each species using a mixed modelling approach and were then correlated to a range of local and regional hydrological and atmospheric time series data at seasonal and annual temporal scales. Biochronologies of 15 to 36 years were obtained and spanned a period of severe drought known as the Millennium drought. All chronologies showed considerable inter-annual variation in growth. Among species, environmental drivers of variation in growth differed. River flow was found to be most influential in driving *M. ambigua* growth in a linear manner, while the growth of *A. japonicus* and *A. butcheri* responded in a linear fashion to fluctuating sea surface temperatures and local rainfall, respectively. These results suggest that generalisations about environmental influences on ecosystem functioning may mask species-specific responses, as fish respond to environmental change dependent on the constraints of their life history strategies. These findings more broadly highlight the importance of considering the diversity of ecological groups that inhabit an ecosystem when developing conservation and management strategies.

Abstract reference: IIA_Morrongiello_05

A 100-year assessment of biological change in SE Australian waters: novel insight using fish otoliths

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Understanding and predicting the impacts of environmental change on biological systems are key tasks facing researchers today. This requires good data. Unfortunately long-term biological datasets are uncommon for aquatic systems and animals such as large-bodied fishes are unsuitable for experimentation. Aquatic biochronologies, generated from time-dependent information recorded in

fish otoliths that are archived in their millions worldwide, can provide valuable long-term datasets that facilitate the development of ecological and evolutionary insights into marine and freshwater environments. South east Australian waters support both unique biodiversity and major commercial fisheries, but the region and its natural resources are increasingly being exposed to rapid oceanic warming. Here I present the results of a large-scale project investigating the environmental drivers of fish growth variation using a data set of unprecedented spatial, temporal and biological coverage. Over 30 otolith-based growth time series, up to 100 years in length, from across nearly 3000km of ocean and a range of habitats have been analysed. Long-term growth patterns for many species display strong temporal synchrony, pointing to universal ecosystem drivers. Directional trends are indicative of warming (via direct and indirect pathways) either promoting or inhibiting growth, whilst quasi decadal oscillations reemphasise the importance of zonal westerly winds in driving recruitment and system productivity variation. Within species, significant growth plasticity exists that may confer some resilience on species faced with climatic change. Together, these results highlight the valuable information stored within otoliths and their potential to provide unprecedented levels of spatial and temporal detail in aquatic environments.

Abstract reference: IIA_Chang_06

Prey composition analysis using otolith morphology: a case study of bigeye tuna *Thunnus obesus* in the western Indian Ocean

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Prey composition of the bigeye tuna (*Thunnus obesus*), an economically crucial fish and an apex predator of the ocean, was analyzed by using otolith morphology. A total of 183 bigeye tuna stomach samples were collected between April 2006 and December 2006 by three Taiwanese longline vessels in the western Indian Ocean. A total of 1,021 free otoliths were extracted from 78 stomachs for identification based on morphology. The results revealed that 49.46% of the otoliths belonged to the family Myctophidae and 21.45% belonged to the family Scopelarchidae, followed by the families Diretmidae (7.54%), Microstomatidae (5.48%), Paralepididae (5.48%), and Macrouridae (4.60%). Three newly recorded prey items, *Valenciennellus tripunctulatus* (Sternoptychidae), *Evermannella* sp. (Evermannellidae), and *Zenion* sp. (Zenionidae) were discovered. The remaining items (e.g., the myctopids *Electrona risso* and *Diaphus* sp., and the paralepidids *Macroparalepis*, *Magnisudis*, *Paralepis*, and *Sudis*) were ascribed to a taxon lower than that of the items observed using the conventional examination of stomach contents. In addition, the effects of acid erosion were evaluated to determine morphological changes in otoliths caused by digestion; certain *Diaphus* specimens were observed to be less durable against acid erosion than other specimens were. The samples did not differ by catch location or the time when they were caught. However, diet was related to growth. Small bigeye tuna preyed mainly on Myctophidae, but consumed increasing proportions of Scopelarchidae, Paralepididae, and Macrouridae, and a decreasing proportion of Myctophidae as the bigeye tuna increased in size.

The use of otoliths to examine trophic interactions between California sea lions and artisanal fisheries on the west coast of the Baja California peninsula

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Some 17 California sea lion (*Zalophus californianus*) rookeries and hauling areas are distributed on islands along the west coast of the Baja California peninsula. The waters surrounding these islands are popular artisanal fishing spots as well. These modest fishing communities can incur significant economic losses as the result of interactions with sea lions (*i.e.*, due to catch loss and damage to fishing gear as a result of depredation); meanwhile, incidental catches can have a detrimental effect on local sea lion populations. While these types of interactions are commonplace, this is the first study to specifically address such interactions off the west coast of the Baja California peninsula. We examined the trophic overlap between sea lions from the Cabo San Lazaro hauling area and artisanal catches by fisherman living in the same area. Sea lion dietary composition was determined through visual identification of fish otoliths recovered from scat. The results were then compared with data on fishery landings, using the Colwell and Futuyama index to assess the degree of trophic overlap. The fish species most commonly consumed by local sea lion populations—*Synodus lucioceps*, *Prionotus stephanophrys*, *Porichthys myriaster*, and *Porichthys notatus*—have no commercial value. These preliminary findings suggest that the degree of trophic overlap between California sea lions and artisanal fisheries in Cabo San Lazaro is quite low. Thus, the alleged depredation of catches by sea lions and the high rate of sea lion entanglement in the area may be due to the spatial proximity between local fishing hot spots and sea lion colonies. This spatial overlap should be examined further.

Parallel Session IIB (Evolution)

Abstract reference: IIB_Tuset_01

May otolith morphology be used for measuring biodiversity of marine fish assemblages?

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In palaeontology, the knowledge of fossil fish assemblages is performed from skeletons and teeth and otolith characters. However, fossil otoliths, especially the sagitta, represent the most common remains of teleost fishes. In this study, we examined if the morphological diversity of sagittal otoliths may be useful as a measure of biodiversity, especially of the functional diversity, since this otic structure is related to audition, mechano-reception and equilibration functions. We also analysed if the body pattern of fishes are linked to otolith shapes, and hence if it is possible to infer morphologies of fossil fishes. Three current coastal fish assemblages of northwestern Mediterranean with different functional diversity and body shapes known were selected to test these hypotheses. The otolith shape was digitalized from 12 landmarks defining the contour features, and others 12 landmarks drawing the sulcus acustics. The results revealed that the fish assemblage with higher functional diversity also showed a greater morphological richness in otolith and body shapes. However, the species distribution within morphospaces using otolith or body shapes was different. Singular otolith shapes were located in the periphery of morphospaces, similarly to species with special morphology of body as *Zeus faber*. Moreover, sound producer specialists as *Sciaena umbra* were also found in the morphospace periphery. It highlights that flatfishes presented a high variety of otolith shapes versus morphological similarity in body pattern. We think that this variability is a good reflection of different ecological strategies among species. Therefore, results suggest that otoliths are adequate for studying the morphological and functional diversity in fish assemblages.

Abstract reference: IIB_Reichenbacher_02

Inter-population differences in otolith morphology are genetically encoded in *Aphanius fasciatus* (Cyprinodontiformes, killifishes)

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Inter-population differences in otolith shape, morphology or chemistry have been used effectively as indicators for stock assessment or for recognizing environmental adaptation in fishes. However, the precise parameters that affect otolith morphology remain incompletely understood. Here we provide the first direct support for the hypothesis, that inter-population differences in otolith morphology are genetically encoded. The study is based on otolith morphology and two mitochondrial markers (D-loop, 16S rRNA) of three natural populations of *Aphanius fasciatus* (Teleostei: Cyprinodontidae) from Southeast Tunisia. Otolith and genetic data yielded congruent tree topologies. Divergence of populations likely results from isolation events in the course of the Pleistocene sea level drops. We propose that otolith morphology is a valuable tool in resolving genetic diversity also within other teleost species, which may be important for ecosystem management and conservation of genetic diversity. As reconstructions of ancient teleost fish faunas are often solely based on fossil otoliths, our discoveries may also lead to a new approach for research in palaeontology.

Abstract reference: IIB_Gaemers_03

On the evolution, systematics and distribution of trisopterine Gadidae

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New data on Tertiary, Quaternary and recent otoliths of trisopterine Gadidae reveal that their evolution and systematics are more complicated than earlier expected. The origin of the two sister species *Trisopterus luscus* and *T. capelanus* most likely is in the Early Miocene. Holocene otoliths from the Ria de Arosa, Galicia, Spain, are additional proof that *T. capelanus* is a good, separate species. Fish morphology and otoliths of *T. esmarkii* are the most aberrant of all species usually included in *Trisopterus*; this species is descended from the older lineage of "*Neocolliolus*" *vikingensis* Gaemers, 1987, and must be placed in the fossil genus *Neocolliolus* Gaemers, 1976. *Trisopterus minutus* is more related to *N. esmarkii*, as genetic data also suggest (Gonzalez *et al.*, 2012), but these are no sister species. *T. minutus* is descended from another lineage than the tree other species and must therefore be placed in a new genus, also because of some different otolith characteristics. The common ancestry of *T. minutus* and *T. luscus* + *T. capelanus* is in the Late Oligocene or perhaps even in the Early Oligocene.

The fossil otoliths demonstrate that extant genetic and morphologic data of recent species are not enough to unravel all relationships between trisopterine species. The paleontological data appear to be crucial for this.

Abstract reference: IIB_Jones_04

Variability in the Santa Barbara Basin Fish Assemblage in the Last Two Millennia Inferred from the Fossil Otolith Record

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The long-term variability of the entire fish assemblage, especially mesopelagic fish, off the coast of Southern California remains largely unknown. The Santa Barbara Basin (SBB) fish assemblage is examined using 1,452 otoliths recovered from four sediment cores with varved chronologies from 33 to 2009 AD. We use otoliths rather than other fish remains to reconstruct the fish assemblage because otolith shape is species specific and the number of otoliths per fish is constant. Otoliths were classified to taxa using geometric and Fourier shape analyses and by direct comparison with references. Otolith elemental composition is also being explored for otolith classification. Our results indicate that mesopelagic, not pelagic, fish dominate the forage fish assemblage and that, like pelagic species, they fluctuate in abundance on decadal time scales. It is also observed that the otolith deposition rate of the taxon bathylagidae is highest in the last 150 years, consistent with recent warming.

Abstract reference: IIB_Lin_05

Fish otolith assemblages on the Recent NE Atlantic sea bottoms

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Reconstruction of paleoenvironment using otolith assemblage has been applied on many marine Cenozoic sediments. However, the compositions of otolith assemblage on Recent sea bottom in various environmental settings are poorly known. Given that they could provide precise reference information while interpreting fossil otolith materials in the reconstruction of paleoecology. This study aims to understand the characters of otolith thanatocoenosis from Recent sea bottom and comparing them with their biocoenosis. Otolith assemblages from box corers or Van Veen samplers collected on the North-eastern Atlantic Recent sea bottom, at various depths and at various latitudes, were analysed. We hypothesize that the bathymetric and biogeographic information interpreted from the otolith thanatocoenosis is reflective of the *in situ* biocoenosis. The results of quantitative analysis indicate that the majority of otoliths enter the sediment at upper part of the slope. Otolith size-related distributions along the water depth are discovered among *Benthosema*

glaciale, *Lampanyctus crocodilus* and *Scopelogadus beanii*, with the increase proportion of larger specimens in deeper waters, which could also imply distribution differentiations in their population structures in the water column. The results of bathymetric method obtained from the otolith assemblage species composition are compared with the actual sampling water depths to test the reliability of this methodology. The bathymetry is improved when only the benthic-benthopelagic taxa are considered. The identified taxa of the assemblages correspond well with the recent biogeographic distribution. Therefore, information such as bathymetry and biogeography from the otolith-based species in the sediments may be used as useful tools in paleoenvironmental reconstructions.

Parallel Session IIC (Ecology)

Abstract reference: IIC_Oehm_01

Determining the foraging grounds of the Great Cormorant by otolith shape and chemistry

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Great Cormorants (*Phalacrocorax carbo sinensis*) are one of the main fish-eating birds hunting in lakes and streams of the Alpine foreland. Initially, they were only overwintering in this region, nowadays they have also established breeding colonies. As a result, freshwater fish must have faced increased perennial predation by cormorants. However, it is poorly known if the birds switch between different feeding grounds and how cormorant prey choice changes throughout the year. This study is the first one investigating this question by characterising and provenancing otoliths of prey fish from cormorant pellets in freshwaters by (1) establishing a reference database for morphological hard part characterisation and (2) strontium isotopic analysis. During breeding season and winter roost regurgitated pellets of the birds were collected fortnightly for two years at lake Chiemsee (Bavaria, Germany). Prey remains were identified using morphological hard part analysis. Differences in water chemistry between different potential feeding grounds and resulting reflection of this information in otoliths (elemental pattern and ⁸⁷Sr/⁸⁶Sr isotope ratio) were evaluated by analysing water samples and otoliths from reference fish collected from lake Chiemsee and its surrounding water bodies using inductively coupled plasma-mass spectrometry (ICP-MS). Additionally, species-specific differences in otolith chemistry of fish from the same site were investigated. Furthermore, otolith shape analysis was performed to examine differences in shape between different water bodies.

The reference data can now be used to classify the otoliths from cormorant pellets according to their origin, and, consequently, for determining the foraging range of cormorants in the Alpine foreland.

Abstract reference: IIC_Landaeta_02

The use of otolith microstructure for analyzing the effects of ectoparasites over nearshore fish larvae

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Most research about ecological interactions between ectoparasites and their hosts has been carried out in the adults and/or juvenile stages of fishes, but there is scarce information related to ectoparasites on fish larvae. These hosts are more vulnerable to the effects of parasites than adult due to their fragile nature and considering they are in early stages of immunological and physiological development. Under this scenario, we hypothesize that ectoparasites over fish larvae will affect the growth and condition of the host, and that effect will be reflected in the otolith microstructure of larvae. The objectives of this study were to compare univariate measurements of sagittal otoliths of parasitized (P) and non-parasitized (NP) fish larvae from two species with pelagic larvae and benthic adults, triplefin *Helcogrammoides cunninghami* (Tripterygiidae) and clingfish *Gobiesox marmoratus* (Gobiesocidae); to compare population larval growth of both species between P and NP larvae by using otolith microstructure analysis, and to estimate recent growth indices of P and NP larvae through microincrement widths. During austral spring 2012-2013 fish larvae were collected in nearshore (< 0.05) than those of NP larvae and they were smaller-at-age and had narrower microincrements than NP larvae. However, in the case of larval clingfish, size of the sagitta was the same for NP and P larvae (one-way ANCOVA, $P > 0.05$), as well as the estimated growth rates (one-way ANCOVA, $P = 0.68$). We discuss the use of otolith microstructure analysis in order to establish variability of parasite holding resistance among larval fish species and variations in larval growth and condition of nearshore cryptic marine fishes.

Abstract reference: IIC_Chung_03

Functional diversity and behavioral responses to depth gradients revealed by otolith morphology in deep-sea fishes

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Fish have diverse otolith morphology, but the connection between otolith shape, otolith function and behavioural ecology is not well known. In this study, otolith shape was analyzed in 898 otoliths from 39 species of deep-sea fishes and used to classify species into distinct groups that correspond to known differences in behavioral ecology. Two further indices associated with sensory function were measured: sulcus area/ otolith area (SO ratio) and otolith mass. Sensory function indices reflected different auditory and vestibular adaptations between the groups classified by otolith shape. Between-species variations in otolith mass were regulated by phylogeny, whereas variations in sulcus area were ecologically determined. Sulcus area ratio decreased with increasing habitat depth, and this trend was stronger in benthopelagic compared to benthic fish. The observed trends are consistent with light-dependent reductions in adaptive pressure for visual hunting in pelagic foraging demersal fishes. Otolith morphology is a powerful record of sensory adaptations that may reveal ecological and evolutionary trends in both modern and fossil species.

Discriminating cryptic speciation using multivariate analysis of otolith morphometrics

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Many factors may contribute toward difficulties in visually distinguishing cryptic sympatric fish species. Phenotypic expressions may be relatively indistinguishable, but advances in molecular techniques have helped to redefine existing taxonomic classifications, resulting in synonyms or resurrections among cryptic species. Alternatively, slight variations in phenotypic expressions in some species may only represent regional variation, rather than speciation. In routine fisheries sampling, fish are often donated from commercial or recreational catches with fillets removed (i.e. lateral skin and flesh from the body) and in a degenerated condition, rendering many diagnostic characteristics absent or indeterminate. Thus, examples of cryptic speciation may require a re-evaluation of species identification post-sampling, particularly for fisheries assessment purposes. To overcome such identification issues, we developed a rapid and reliable method for distinguishing between two phenotypically similar Eteline snappers (*i.e. Etelis carbunculus* and *E. marshi*) using simple otolith morphometrics (length, width, thickness and weight) with or without fish length, within a traditional canonical discriminant analysis (CDA). This method achieved a very high allocation success rate using either otolith morphometry and fork length (99.6% for *E. carbunculus* and 100% for *E. marshi*) or otolith morphometry only (98.8% for *E. carbunculus* and 100% for *E. marshi*). The CDA successfully grouped samples of the same species across a wide geographic range from numerous locations throughout the eastern Indian and western central Pacific Oceans. We are now using the method with multiple species in other family complexes to test its broader applicability. This method may also have broader application for species identification involving skeletal or external anatomic morphometrics in extractive, diet or video based studies.

Poster Session (IIE)

Abstract reference: IIE_QUIJANO-PUERTO_01

Age and growth of the invasive lionfish (*Pterois volitans*) in the Parque Nacional Arrecife Alacranes, Southern Gulf of Mexico

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The lionfish (*Pterois volitans*), a native reef fish to the Indo-Pacific Ocean, was introduced in the Atlantic Ocean 30 years ago where it has now invaded a great portion of this region, and represents an environmental threat because of its population growth. In the Southern Gulf of Mexico, off the northern coast of the Yucatan Peninsula, specifically in the Parque Nacional Arrecife Alacranes (PNAA), the lionfish invasion is at an advanced level; however, there have not been studies of lionfish's corporal growth in the area. Consequently, the objective of this work was to determine its age and growth, as determined by using sagittal otoliths, and calculate both the Von Bertalanffy and Gompertz growth functions. From 2010 to 2012, about 800 lionfish (97 mm to 384 mm TL) were collected from which 609 whole sagittal otoliths were removed and prepared for reading. Preliminary data would be presented in this work, which represents a baseline for future studies on lionfish's population dynamics to understand the invasive process in the region.

Abstract reference: IIE_MAHOUACHI_02

Application of otolith shape analysis in identifying different species of *Zosterisessor ophiocephalus* and *Gobius paganellus* in the lagoon of Bizerte in Tunisia

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The two species of gobies *Gobius paganellus* (Linnaeus, 1758) and *Zosterisessor ophiocephalus* (Pallas, 1811), are very common in Mediterranean waters, they are easily identified by their pelvic fins which are usually welded along their entire length.

The analysis of otolith shape based on elliptic Fourier analysis (A.F.D.) followed by a statistical analysis is an excellent tool for discrimination of families, genus, species and even fish populations of the same species. In this context, the present study is to separate two species of gobies mentioned, living in sympatry in the Bizerte lagoon (northern Tunisia).

The results highlight the presence of net morphological differences between left and right otoliths of both species. Population of *Z. ophiocephalus* is located on the positive side of the axis F1 (absorption = 61.94%) and the population of *G. paganellus* is located on the negative side of the

same axis F1. While F2 axis (absorption = 24.37%) separate, right and left otoliths and those of *Z. ophiocephalus* and *G. paganellus*.

Own calculated value is less than the significance level (p-value <0.05), or the presence of significant differences between these two populations. So we can conclude that there is no resemblance between these two populations living in the same lagoon environment.

Abstract reference: IIE_Curin-Osorio_03

Comparative morphometry of the sagitta otolith in *Sardina Común (Strangomera bentincki)* and *Sardina austral (Sprattus fuegensis)* in southern Chile

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The morphometry of sagitta otoliths of two species of clupeiforms was compared, the common sardine (*Strangomera bentincki*) and southern sardine (*Sprattus fuegensis*). Both species are pelagic and planktivorous, and they are distributed in southern waters off Chile. In addition, the species can be easily confused because they are very similar in their body morphology and external characters. With the aim of providing criteria for identification of both species, traditional and geometric morphometry techniques were applied to compare the sagitta otoliths. It is postulated that size differences of sagitta otoliths (length, breadth, perimeter and area), shape-based descriptors such as circularity (CI), rectangularity (RI), and ellipticity (EI) indices, and outline analysis based on elliptical Fourier transformed could help to identify each species. The multivariate analysis for length, breadth, perimeter and area, CI, RI and EI as well as for normalized elliptical harmonic coefficients (An, Bn, Cn, Dn) showed significant differences between sagitta otoliths of *S. bentincki* and *S. fueguensis*

Abstract reference: IIE_Brind'Amour_12

Combining biological markers to estimate the realized niche species: the case of three flatfish in the Seine estuary

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Habitat fragmentation and habitat loss have been shown to cause a significant decline in fish productivity, involving the replacement of areas of high productivity by less productive areas. Identification of such high-quality habitats requires however appropriate information on species spatial distribution. Most of species habitat models are developed using abundance data, relying on snap-shot information indicating where a species is located on a given sampling date. This type of information implicitly hypothesize that these snap-shot abundance are characteristic of the species "realized niche": A species realized niche being thought of as the range of habitat types from which a species is not excluded by competing species. One could however argue that developing habitat models using information integrating the life history of a species may be more relevant than using snap-shot data. This study aims to develop a generic method to combine biological markers

integrating information on the life history of species in the seine estuary (stomach contents, muscle isotopic compositions, otolith microchemistry), and compare habitat models developed for three flatfish (sole, solenette, plaice) using the markers index and the conventional abundance data. Results underline some spatial correlation between the two indices, with Pearson r coefficient ranging between 0.43 and 0.62 (for the sole and plaice respectively). The spatial distribution of the markers index being enclosed within the one of the abundance data for the three species. Habitat models developed using the markers emphasize similar realized niche for the sole and plaice, with a high relative contribution of depth and the energetic content of their prey.

Abstract reference: IIE_Sousa_13

Ecomorphological patterns of the sagittae otolith in tropical fish of the western south Atlantic

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The morphology and morphometry of sagittae otolith were studied in tropical marine fish associated with different mobility guild. The shape, margins and rostrum of 396 sagittae from 49 species were classified in: group 1 (migrants marine fish N=15 species), group 2 (vagrants marine fish, N=28 species) and group 3 (territorial fish, N=6 species). The value of E=maximum width of the sagitta (WO)/maximum length of the sagitta (LO%), expresses the relative tendency in the shape otolith (from circular to elongate). The value of R=length of the rostrum (LR)/LO%, expresses the percentage in the total length of the otolith that corresponds to the rostrum were calculated for each species. The sagittae of group 1 were fusiform, oblong, elliptical and square with rounded borders, the rostrum can be absent and the values indices were $E=62\pm 14.6$ and $R=27\pm 7.8$. The sagittae shape of group 2 were elongated with rostrum well developed, leaf-shape with irregular and crenulate posterior margin and rostrum absent, oblong without rostrum, fusiform and globular without rostrum, the value mean were $E=51\pm 14.3$ and $R=34.4\pm 41.2$. The sagittae of group 3 possessed an oblong form, with rostrum poorly developed or absent, the mean values of the indices were $E=66\pm 10.2$ and $R=5.4\pm 2.8$. We found significant differences in the E and R index between the groups ($F(2, 391)=8,56$; $p=0,00023$). The E index was higher in group 3, while the R index was the lowest. Our results suggests that the sagittae otolith in fish of the Carangidae and Paralichthyidae families present several morphological variations associated with the body shape and the substrate type, although are classified in the same mobility guild. The E and R index could be considered a useful tool for fish ecology studies.

Abstract reference: IIE_Tuset_13

Sagittal otolith morphology helps to explain the invasion success of Lessepsian species

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Lessepsian species are marine organisms that enter the Mediterranean from the Red Sea through the Suez Canal. This phenomenon seems to have increased dramatically in recent decades. Here we compared the contour of sagittal otoliths of Lessepsian vs native Mediterranean fishes (families: Atherinidae, Mugilidae, Serranidae, Haemulidae, Callionymidae, Mullidae, Sphyraenidae and subfamilies Clupeinae, Serraninae, Caranginae, Gobiinellinae, Scombrinae). The analyses were based on wavelet functions of an intermediate scale (WLT 5) of 512 cartesian points. A principal components analysis was used to build a morphospace for each group studied. Lessepsian otoliths were characterized by a high morphological differentiation with respect to native ones and tended to be allocated out of the native morphospace. In previous studies we observed that the percentage of correct classifications of Lessepsian otoliths (92.5%) was greater than that obtained by using native species (72%). In some cases (*e.g.* Plotosidae and Fistularidae), the high morphological distinctness of the Lessepsian otoliths can be attributed to phylogenetic distances between these species and the receiving community. Nevertheless differentiated otolith shapes were also observed in those invaders that are phylogenetically related to native species. This peculiar morphology of Lessepsian otoliths within the Mediterranean context might reflect novel functional and behavioural adaptations of these species and hence novel opportunities to thrive in their newly colonized environment.

Abstract reference: IIE_Lampart-Kaluzniacka_14

Effect of disturbances in the structure of trophic lakes on the formation of annual marks in predatory and planktivorous fish

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The studies focused on age assessment based on hard structures otoliths and dorsal fin rays in planktivorous and predatory fish from Lake Trzeciecko (NW Poland). This reservoir is subject to reclamation treatments: aeration of the deep water, precipitation of phosphorus and biomanipulation. Planktivorous fish included roach *Rutilus rutilus*, bream *Abramis brama* and silver bream *Blicca bjoerkna* while predatory fish included pike *Esox lucius*, zander *Sander lucioperca* and perch *Perca fluviatilis*.

It was determined that a very high correlation relationship exists between the length and weight of otoliths and the length of the individual fish. At the same time it was noted that the age of predatory fish were in the range 2+ - 3+ years for zander and pike and 2+ - 8+ years for perch.

Planktivorous fish were significantly older. Their ages were estimated in the range 4+ - 8+ years for bream, 6+ - 7+ years for silver bream and 6+ - 10+ years for roach.

In the case of planktivorous fish very clear artifacts appear on the otolith sections, highly correlating with periods of coagulant (ferrous sulfate) application, the precipitating particles of inorganic phosphorus and plankton, which is the main source of food for these organisms.

These artifacts could, due to lack of knowledge about the environment, overstate the age of the fish examined. The designation of annual marks for planktivorous fish was made possible by microscopic preparations of dorsal fin rays where the boundary and the number of rays was clear. Otoliths proved to be very sensitive structures, closely reflecting the environmental situation prevailing in this reservoir. In this case, it was related to the absence of food as demonstrated via additional lines inhibiting growth. In predatory fish, there were no such artifacts.

Abstract reference: IIE_Callicó Fortunato_15

Identification of Mediterranean Mulletts using morphology of sagittae otoliths

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In the Northeastern Atlantic and Mediterranean Sea there are 7 well-established species of the Mugilidae family: *Mugil cephalus*, *Liza aurata*, *Liza ramada*, *Oedalechilus labeo*, *Chelon labrosus*, *Liza saliens*, *Liza carinata*. The differentiation of mugilids is very important for local fisheries management and regulations, but their identification using external morphological characters remains very difficult. In this contribution, we use the saccular otolith features of each mugilid species to identify these seven common species present in the Mediterranean Sea. We found a general Mugilidae pattern in the otolith morphology and specific characteristics for each studied mullet species could be recognized. An identification key using sagittae otoliths was developed. The results could be useful as a reinforcement of the present identification keys that differentiate through morphological and meristic characters; and also to identify those species consumed by piscivores, being the otoliths the only identifiable remains of the individuals.

Abstract reference: IIE_Durieux_17

Insights on otolith shape diversity of Mediterranean postlarval fishes

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Teleosts are the most diverse Vertebrates group on Earth and it is known since the end of the Nineteenth century that each species display specific otolith shape and size. Otoliths are present in the internal hear of Teleostean fish from the embryonic stage onwards, yet there is very poor information on otolith shapes of fish early life stages and their ontogenetic changes till the adult

stage. Postlarval stage is a transition stage between larval and juvenile stages where individuals change drastically in terms of morphology, so that taxonomic identification can be particularly difficult. In relation to the fishes' post-larvae biodiversity monitoring of Northwest Mediterranean (Life+ SUBLIMO), the aims of this study were: i) to characterize otolith shape of several taxa of postlarval Mediterranean fishes, ii) to evaluate the potential changes of otolith shape between postlarval and adult stages and iii) to define the use of otolith shape as an identification criterion in postlarval fishes. Postlarvae were collected at night using light traps along the Corsican coast in 2012 and 2013. Individuals (n= 166 belonging to 12 taxa) were first identified based on external morphology and meristics before otolith extraction. Sagittal otoliths were then digitized under a stereoscope; shape analysis was performed on contour shape using Fourier analysis and geometric morphometrics methods. In addition, the mean difference between postlarval and adult consensus shape is calculated in order to compare the ontogenetic changes between taxa. To complete the entire surface shape description of otoliths, Scanning Electron Microscope photographs are presented for each taxa. Discriminant Function Analysis showed very high correct reclassification rate (over 90%) of postlarval taxa based on otolith shape analysis. This study provides basic description of the otolith shape of several Mediterranean fishes at the postlarval stage while highlighting its use as a complementary identification criterion.

Abstract reference: IIE_Gerard_18

Isotopic signatures in the otoliths of reef-associated fishes of southern Florida: linkages between nursery grounds and coral reefs

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Ecologically and economically important coral reef fishes are believed to migrate to reefs from juvenile nursery areas such as sea grass and mangrove habitats. Little is known about the migration corridors that exist between nursery and coral reefs, or the timing of these migrations. This study investigated the possibility of identifying connections between juvenile nursery areas and nearby coral reef adult habitats using stable isotope analysis of the otoliths of Gray Snapper (*Lutjanus griseus*) (n= 71) and Yellowtail Snapper (*Ocyurus chrysurus*) (n=60), collected from 9 sites along the Florida Keys coral reef tract in 2003. Measurements of carbon and oxygen isotope ratios were compared to existing isotopic signatures of juvenile snapper from nursery areas around southern Florida. Data showed overlap for ¹³C and ¹⁸O isotope measurements for the juvenile portion of adult otoliths to isotope values for the middle and lower Florida Keys regions, thereby suggesting a migratory connection. Specific nursery habitats contributed approximately 93 % of individuals sampled for Yellowtail and approximately 98 % for Gray Snapper to the Florida Keys coral reef tract. Results from this study indicate locally important source regions and habitats for the successful recruitment of fish to the Florida Keys coral reef tract.

Abstract reference: IIE_Shen_19

Migration history comparison among three *Mugil cephalus* cryptic species in Northwest Pacific revealed from otolith Sr/Ca ratio

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Mugil cephalus is an estuary dependent species during their growth stage and would migrate to the marine environment for spawning. However, *Mugil cephalus* in Northwest Pacific is not only one species, but three cryptic species (NorthWest Pacific1, NWP2, NWP3). These three highly divergent mitochondrial lineages exist in sympatry in Taiwan. Do they have different migration history when living in different area? or have their own species-specific migratory behaviour is still unknown. Through the analysis of their otolith microchemistry in the same or different areas, we can understand more about their habitat selection, migratory behaviour and the evolution mechanism. In this study, the three mullet species in Northwest Pacific were collected from south Taiwan (Pingdon), south China (Hainan Island) and north China (Tianjin). The otolith Sr/Ca ratios were

analysed for different species from different areas. The results suggested that different species may have facultative migratory behaviour according to the local environment. NWP2 and NWP3 prefer to stay in freshwater in Taiwan. However, the same species in Hainan Island seldom got into freshwater. NWP1 prefer to stay in estuary and marine environment in Taiwan but the same species from Tianjin were stay in both marine and freshwater environments. It suggested that the habitat selection for different mullets might be environment dependent.

Abstract reference: IIE_Gholami_20

Morphology of the sacular otoliths in species identification, phylogeny and systematic of the Iranian *Aphanius* (Cyprinodontidae; Cyprinodontidae) species

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The Iranian plateau keeps the highest diversity of *Aphanius* species, with 14 species that represent three large clades based on the cytochrome b gene, *i.e.* the *A. dispar* clade, *A. mento* clade and the Iranian inland and inland-related *Aphanius* species (IIRAS) clade. The species of the *A. dispar* and *A. mento* clades can be identified based on both molecular data, otolith data and external characters, but species identification based on color patterns or external morphological characters is not always possible for the species of the IIRAS clade, which consists of ten species. However, molecular data is not always available. In that case, the morphology and morphometry of the saccular otolith (sagitta) represent a useful tool for a reliable taxonomic identification of the species of the three clades. Key characters of the otolith morphology include the sulcus shape and mineralization, the dimensions of the rostrum and antirostrum, the size of the excisura angle and the overall shape. We found that three basic types of sulcus can be defined that may show the evolutionary sequences of the 14 Iranian *Aphanius* species. Additionally, three types of development in rostrum and antirostrum dimensions appear as diagnostic characters for the species of the IIRAS clade. Furthermore, also otolith morphometry is useful for species identification as the canonical discriminant analysis (CDA) with jackknifed cross-validation revealed a good to high overall species classification success (68–85%) for the species of the IIRAS clade. It is therefore concluded that combination of qualitative characters (sulcus morphology, rostrum and antirostrum dimensions, excisura size) with quantitative approaches (otolith morphometry) represents a powerful method to identify *Aphanius* species from Iran and thus can lead to a better understanding of their taxonomy, diversity and zoogeography. The results are also important for paleontological studies because otoliths are often the sole fossils of ancient fish faunas and are significant to reconstruct ancient fish diversity, zoogeography and evolution.

Abstract reference: IIE_Godiksen_21

Otoliths as life history indicators

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The Institute of Marine Research routinely collects about 80.000 otoliths annually from surveys and reference fleet. For some species, like cod, the collections began in the early 1900s and cover many of the fluctuations in population dynamics seen in the commercial fishery. Age estimation is often done on board soon after catch, for management purposes, but more recently this extensive archive is being accessed with multiple analytical techniques for ecology and climate studies. The IMR archive offers a rare window on past populations and ecosystems and exploitation patterns in high latitudes and the arctic. These otoliths are being used as: Community indicators: - Trophic history of cod over a 100 year period using stable C and N isotopes from otolith protein Population indicators: - a DNA source to track the genetic basis of fishery-induced evolutionary changes in the Barents Sea cod and population structure of hake - Otoliths, spines and vertebra are used in validation of age readings for assessment of the species Individual indicators: - Spawning zones and evidence of skipped spawning in saithe - Validation of first annulus by daily growth increment analysis in hake - Validation of structures in OTC marked and recaptured of Greenland halibut Environmental indicators: - Shape analysis to distinguish herring from different areas, North East Arctic cod from Coastal cod, and wild from farmed cod - Temporal and spatial differences in larval herring growth and survival linked to environmental conditions, available food and predation - The history of heavy metal contamination, especially mercury, recorded in otoliths of Greenland halibut, burbot, and other demersal species.

Abstract reference: IIE_Wang_22

Spatial-temporal distribution and migratory pattern among mullet species in Tanshui River

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For investigating the mullet species composition and migration pattern among species, a total of 848 mullet were collected every season from Tanshui River. Species were identified by previous studies that using mtDNA to classify 3 cryptic species, NWP1-3. Otoliths were aged and VBGE were estimated. Polished otolith were followed by micromilling to sample each annuli for elemental analyses to investigate the ontogenetic migration behavior of each mullet species. The most abundant mullet species in Tanshui River is NWP2, compositing 66.1, 74.6 and 97.1% of all specimens in winter, spring, and summer, while seasonal composition of NWP1 is 32.3, 23.8, 2.9 % consequently. Only in autumn, NWP1 rose up to 60 % and was higher than NWP2 (32%). Meanwhile, NWP3 remained consistently lower than 10 % throughout the year. In autumn, GSI of

NWP2 is higher than NWP1, indicating that NWP2 individuals migrated to open ocean for spawning and the percentage of NWP2 remaining in the river therefore lowered down. For spatial distribution, only large and older mullet individuals inhabited at freshwater upper reaches and the pattern showed similar in all species, indicating that only older mullet can stay in absolute freshwater environment, while juveniles and young mullet stayed in saline water with higher productivity and more abundant food. Results of otolith trace element signature in both core and edge all showed significant different, implying existing different spawning ground and migratory pattern among species. In addition, the trace element signature of NWP1 is similar to main wild spawning mullets in Taiwan Strait. Otolith edge elemental signature is efficient for distinguishing the marine and riverine individuals, but the discrimination efficiency is lower between locations in Tanshui River system.

Slideshow Poster Session (IISS)

Abstract reference: IISS_Chang_01

Using otolith morphology in zoogeography and phylogeny analyses of Sciaenidae

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Sciaenidae is characterized by having a pair of distinctly large and often thick otoliths. Morphology of sagittal otolith, its shape, thickness and sulcus mark on the inner surface are characteristic at generic or specific levels, which are widely applied in systematics, fishery biology, trophic ecology, paleontology and archeology studies. However, these morphological characters have not been systematically listed except in some otolith atlases. In total, 432 records attributed to 64 genera and 195 species of the sciaenids were documented worldwide. Otoliths of 43 genera and 118 species are currently archived in the NMMBA for the Global Sciaenidae Conservation Network (GSCN). There are unique morphological patterns to characterize sciaenids; some genera have thinner sagittal otoliths, such as the American *Aplidinotus*, *Cynoscion*, and *Menticirrhus*, which the last one has its gas bladder atrophied in adults; one West Pacific species, *Miichthys miiuy*, also has thin otolith. The genera *Johnius* (Indo-Pacific) and *Stelliferini* (Americas) have reduced sagitta and enlarged lapillus. In order to quantify these descriptive patterns, the otolith dimension characteristics (ODC) and Fourier shape description (FSD) are measured and analyzed with the photographs of left sagittal otoliths. The inner surface measurements, perimeter, width/length, angle, circularity, round, solidity and thickness were used for ODC measurements. Multivariate analyses were applied to the discriminant among genera and species to build sciaenid phylogeny systematics. Otolith morphologies can reflect both phylogenetic and environmental patterns. The phylogenetic pattern is significant in distinguishing the sciaenids of three zoogeographic regions, Americas, East Atlantic and Indo-Pacific. Otolith morphology also varies ontogenetically and in response to indicate different habitat use. Using quantified otolith morphology is efficient in genera classification but not always in species. In some genera, the species-specific is significant, such as *Bairdiella*, *Stellifer*, *Ophioscion*, *Larimus* and *Pseudotolithus* and in part of *Cynoscion* and *Johnius*.

Abstract reference: IISS_Chang_02

Fish otolith assemblages from Tainan Science Park archaeological sites in southwestern Taiwan

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Coastal adaptation has been a crucial strategy for prehistoric settlement in Austronesia speaking zone. The archaeological sites located in Tainan Science Park, Southwest Taiwan are on the northern margin of the Austronesia speaking zone, which provide opportunities for exploring early development of the coastal adaptation. This study investigated three archaeological middens, Nan-kwan-li East (NKLE), Niu-niao-kang (NNK), and She-nei (SN), which were dated 4800~4200 BP, 3300~2300 BP, and 450~290 BP, respectively. With the aim of understanding coastal adaptation, the study focused on evaluating fishing ability between these middens using fish otolith remains. Otolith assemblages were identified to reveal the taxon composition. The results show at least 21 taxa were consumed in NKLE (n=580), nine taxa in NNK (n=31), and one taxon in SN (n=36). Sciaenids were the dominant components both in NKLE and NNK, and genus *Larimichthys* was particularly abundant. *Arius* was the only taxon recovered from SN. The majority of shallow water otolith composition with moderate-sized dimensions could indicate a neritic food source. However, several adult epipelagic otolith specimens imply more oceanic demands which pertain to better coastal adaptation.

Abstract reference: IISS_Lombarte_03

Identifying Mediterranean Sea gobies using otoliths

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A total of 234 sagittal otoliths from 25 Mediterranean (North Western area and Adriatic Sea) gobiids species were anatomically described and morphometrically analysed to determine the usefulness and efficiency of sagittae in the identification of species. This group of fishes is one of the largest Mediterranean Teleostean families characterizing by a high taxonomic complexity. The analysis of contour otolith was based on the mathematical descriptor called wavelet (WT). Two methods of classification were used in basis wavelets functions: an iterative system based in 10 wavelets that searches on AFORO database; and a discriminant method based in WT number 5. With exception of paedomorphic species, the results showed that otolith anatomy and morphometry can be used as diagnostic character of the three Mediterranean phylogenetic lineages (*Pomatoschistus*-lineage or sand gobiids, *Aphia*-lineage-gobiids and *Gobius*-lineage). The main anatomical differences of the otoliths were related to general shape (squared to rhomboidal), the development and shape of postero-dorsal and antero-ventral lobes and the degree of convexity of dorsal and ventral margins. In basis to morphometry of otolith contour, iterative classifications and discriminant analysis showed very similar results. In both cases more of 70 % specimens were correctly classified at species level and more of 80% at genus level. Iterations on AFORO database (including 216 families of Teleostean fishes) give a 100% of correct classification at family level. These high differentiated otoliths shape respect other Perciformes families support the most recent molecular data that propose recognition of gobiids and related groups as a new taxonomic order, Gobiiformes, separated of the main Perciformes taxa.

Abstract reference: IISS_Songer_04

Maintaining quality standards throughout the collection, processing, reading and archiving of otoliths

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Cefas' otolith collection is by far our biggest physical data collection, which not only underpins fisheries assessment, but also represents our most valuable unrealised source of retrospective physical and biological data. We are adding to the collection continuously with otoliths being collected from a variety of sources. This poster aims to demonstrate how we maintain good data quality throughout the whole process of adding to this valuable collection and utilising its contents. From collection of samples and associated data using our Electronic Data Capture system through the processing and reading of otoliths in our ISO 17025 accredited lab and on to our archive with its interactive search portal.

Abstract reference: IISS_Khan_05

Otolith research in India: status and perspective

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The conservation and management of aquatic resources become essential for the sustainable harvest of fisheries potential today as well as for future generations. Studies on age and growth, life history, larval dispersal, movement and stock structure are important aspects of research in fish biology. Such information is necessary for the scientific interpretation of the alterations in the fish population and then to develop efficient fisheries management strategies for the target fish species. Some inherent characteristics of otoliths make them excellent marker used in fisheries management. Otoliths (ear stones) are located in the head region and bathed in endolymph within a semi-permeable membrane of the inner ear of all teleost fish and act as a balance organ and also help in hearing. Otoliths are formed during embryogenesis, grow continuously throughout a fish's life, and incorporate both minor and trace elements during growth. Earlier work on otoliths in India was restricted to only age determination by counting the annuli. In recent years, researchers in India have started to explore otolith chemistry to identify population structure, otolith shape for species identification, chemical composition of otolith as pollution indicator and daily growth increments. However, the pace and quality of research on otoliths in India is nowhere comparable to the leading otolith research laboratories of the world. Few research groups have independently started working on modern aspects of otoliths research but at smaller scales. Much more efforts are warranted on otoliths research from India to make its use in understanding and managing the target

fish population better. The present paper is aimed to review the status and future direction of otolith research in India.

Abstract reference: IISS_Mahouachi_06

Otolithometry aspect of differentiation of *Trachurus trachurus* (Perciformes, Carangidae) in the lagoon of Bizerte and the island of Galite in Tunisia

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Horse mackerel (*Trachurus trachurus*) is a teleost belonging to the family Carangidae; which gathers in a pelagic bench on sandy bottoms, but sometimes the depth. This species is experiencing a wide geographical distribution populating the entire Atlantic, the Mediterranean basin including the Black Sea, the Pacific and even the Indian Ocean. Otolithometric analysis is a technique widely used in the differentiation of species of fish and the discrimination of their stock in different environments. The saccular otolith or sagittae is the largest and most in diagnostic research. This work aims to make the differentiate between *Trachurus trachurus* population of the Bizerte's lagoon, located in the extreme north of Tunisia over an area of approximately 128 km² with a maximum width of 11 km and a maximum length 13 km, compared with individuals from the island of la Galite, located in northwest of the Tunisian coast, measuring 5.4 km in length from east to west and up to 2.9 km width in its eastern part, by otolithometric approach to analyze the shape of the left and right otoliths. The results obtained demonstrate the presence of a difference between males and females of both populations. This result allows assuming sexual dimorphism in otolith of *Trachurus trachurus* in the two media studied.

Abstract reference: IISS_Rossi-Wongtschowski_07

Teleostei Fish Otoliths from the S-SE Brazil: a sagittae collection, its website and publications

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This work presents the Collection of Otoliths from teleost fish of the S-SE Brazilian coast - COSS Brasil hosted at the Laboratory of Fish Fauna and Growth (LABIC) of the Oceanographic Institute of the University of São Paulo (IOUSP), Brazil. The collection is composed of more than 45,000 pairs of sagittae from 210 species. A Manager Database System MySQL using an Entity-Relationship Modeling is used to store information on fish and fish otoliths. The system comprises five modules including fish biological data (*i.e.*, length, weight, sex and maturity stage); oceanographic data (*i.e.*, date, sample coordinates, temperature, salinity and weather conditions); taxonomy (*i.e.*, species names, family, order, species descriptions and acronyms); otolith information (*i.e.*, availability, morphological and morphometric data); permission to secure access; and language flexibility. The goal of the LABIC is to be a center for the study of otolith morphology, age and growth and a

depository for the sagittae of Southwest Atlantic fishes, make the collection available to researchers all over the world. At present some results are available in www.usp.br/cossbrasil. We also aim to publish the results in an online practical guide with detailed drawings and photos of otoliths of different sizes in medial and lateral faces, as well as ventral profiles. Statistical analysis to evaluate ontogenetic differences will also be available. There are currently no otolith catalogs for SW Atlantic fish. In order to make these results quickly available, otolith descriptions for 11 species of Gadiformes and 36 of Perciformes were recently published and are available in a special edition of the Brazilian Journal of Oceanography at <http://www.io.usp.br/article678>.

Abstract reference: IISS_Reichenbacher_08

The Late Miocene freshwater fish fauna from Venta del Moro (Valencia, Spain)

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We report on a fossil fish fauna from Late Miocene sediments at the site Venta del Moro (near Valencia, Spain). The site Venta del Moro could be dated based on mammals and magnetostratigraphy and is approximately 5.8-6.2 Ma old. The reconstruction of the fossil fish fauna is exclusively based on the abundant occurrence of exceptionally well preserved otoliths; no skeletal remains have been preserved. Most of the otoliths from Venta del Moro could be identified down to species level and more than 35 species are present. The most speciose groups are killifishes (Cyprinodontiformes) and gobies (Gobioidei), with more than 10 species in each group. Among the killifish otoliths, we found a species closely related to Valencia MYERS, which is the first fossil record of this genus. This fossil Valencia species may be closely related to the Recent *V. hispanica*, which is an endemic and threatened brackish and freshwater species living in the Mediterranean area, near Valencia (Spain). Notably, the fossil freshwater fish fauna from Venta del Moro is more speciose than any previously described freshwater fish fauna from the European Miocene. It can be assumed that a large, probably slightly brackish water body existed at Venta del Moro during the Late Miocene. This large inland water body apparently supported heterogeneous habitats with different water depths, substrates and types of prey. However, the high species diversity in the ancient Venta del Moro inland water body may also have resulted from the prominent climatic and palaeogeographic changes in the Mediterranean area during the Late Miocene.

Theme III: Population Indicators

Plenary session (IIIA)

Abstract reference: IIIA_Thorrold_Key

Applying otolith geochemistry to quantify alternative population attributes needed for ecosystem-based management of marine and freshwater fishes

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Otoliths have provided fish biologists with a treasure trove of valuable life history information across levels of biological organization from individuals to ecosystems. Age and growth estimates have been particularly important in the history of otolith science largely due to their relevance to traditional models of population dynamics that have been the mainstay of fisheries science for more than half a century. I will focus on the path less traveled by describing work in our lab that uses otoliths to estimate population attributes that are not commonly used in traditional fisheries assessment but that have become increasingly important as we transition to spatially explicit and ecosystem-level management tools. For instance, otoliths have proved essential in a number of studies of population connectivity, both through larval dispersal and natal homing and straying. These studies have largely concentrated on the inorganic geochemistry of otolith aragonite and using technologies including laser ablation and multiple collector inductively coupled plasma mass spectrometry. Results have forced us to re-think the spatial scales of population structure in marine fish. We have also developed techniques for quantifying stable carbon and nitrogen isotope ratios from specific amino acids in otolith proteins. Using this approach it is possible to retrospectively determine carbon and nitrogen sources at the base of foodwebs by referencing sampling locations to time stamps in otolith sections determined from increment patterns. Initial results are intriguing as they suggest that it may be possible to reconstruct the diagnostic characteristics of the food web architecture from habitats that fish have used throughout their lifetimes. Taken together, analysis of both the inorganic and organic components of fish otoliths will continue to have numerous uses for fisheries scientists transitioning to ecosystem management in the presence of significant global climate change.

Abstract reference: IIIA_Wright_01

Identifying the mechanisms shaping population structure in fish with dispersive life stages

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Two key mechanisms shape population structuring in the open seas; physical forcing and philopatry. Hydrographic processes may segregate planktonic offspring of different spawning aggregations while behavioural mechanisms may enable individuals to return to the spawning area they

originated from. Few applications of otolith micro-chemistry have investigated philopatry and those that have, have focussed on species with at least one life stage that exhibits high site fidelity. In this study we examined the mechanisms behind population structuring in a more typical marine species where all life-stages can disperse; the Atlantic cod. Samples of juvenile and adult cod from the same year-class were obtained from across the North Sea, Skagerrak and Kattegat. Using a combination of laser ablation and whole solution ICPMS, we tested whether genetic evidence for philopatry was linked to full life-stage fidelity or natal homing and whether finer scale patterns of adult fidelity evident from tagging was related to entrainment following settlement based on comparisons of otolith microchemistry of different life-stages. Regional differences in otolith micro-chemistry were maintained across life-stages consistent with the two genetically differentiated cod populations previously identified within the North Sea. Both hydrographic separation of larvae and a limited north-south dispersal of adult cod were suggested by otolith microchemistry, consistent with biophysical models of cod larval transport and electronic tagging studies. In addition, based on the larval and 0-group component of adult otoliths, the western Skagerrak received larvae spawned in the northern North Sea and adults returned from that area, consistent with natal homing. In some coastal areas, patterns in otolith microchemistry reflecting development up to the settled 0-group suggested an active return migration of settling juveniles. This study demonstrates that it is possible to identify the nature of population structuring even when all life-stages can be dispersive using a variety of otolith microchemistry approaches.

Abstract reference: IIIA_PANFILI_02

After two centuries of fish sclerochronology: the hegemony of otolith studies

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This review evaluates the different steps of discoveries in fish sclerochronology over the time by analyzing the amount of published works in each field of research and applications. Even if the first growth marks in fish calcified structures were evoked during the 18th century, the first serious attempts for estimating the age of vertebrates using sclerochronology were made at the end of the 19th century. At the beginning of the 20th century, scales and otoliths, and more scarcely bones, have been used concomitantly to study seasonal growth cycles. Two major steps allowed specifically the development of otolith studies, the first at the beginning of the 70's with the discovery of daily micro-increments, and the other ten years later with the analysis of microchemical constituents to reconstruct environmental life history. The development of new methods for validating the age estimation through microchemistry rose at the end of the 20th century. Since then, after more than one century of investigations, sclerochronology remains one of the most innovative tool for studying fish biology and ecology. A global literature survey from two major bibliographic web-databases, the Web of ScienceTM and ScienceDirect, gave a clear image of the present hegemony of otolith studies in sclerochronology. During the last decade 80% of studies used otoliths, the remaining 20% using scales and bones, against 40% for otolithometry and 60% for scalimetry or skeletochronology in the 80's. More recently the development of isotope analyses from the otolith protein matrix allowed new investigations on the present and past fish diet. Advanced technologies on otoliths also influenced researches on scales and bones, particularly in paleo-archeology which constituted around 10% of the recent publications. The discoveries in the use of otoliths in ecology are still rising, but until when?

Abstract reference: IIIA_Folkvord_03

Herring year classes after 1904 - how have they affected growth of subsequent year classes?

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The 1904 year class of herring dominated the Norwegian spring spawning herring (NSSH) stock over a decade in terms of numerical abundance and contributions to the fishery, and density dependent effects have been postulated. We compare the growth of large year classes of NSSH with the two subsequent year classes, starting with the strong 1950, 1959, 1983, 1991, 1998, and 2002 year classes. Back-calculated lengths-at-age were estimated from an extensive historic NSSH scale collection. The two year classes following a strong year class typically exhibited higher growth during the pre-recruit phase than the strong year class. An exception to this pattern is seen for the 2003-2004 year classes following the strong 2002 year class. The 1983 year class which was the first large year class after the stock collapse in the mid-sixties had the highest length-at-age 6 of all strong year classes. The large 1950 and 1959 pre-collapse year classes tended to have smaller lengths-at-age than strong year classes after the collapse. The results are discussed in relation to the regional environmental and climatic conditions.

Abstract reference: IIIA_Darnaude_04

Untangling inter-stock differences in otolith d¹⁸O signatures: insights from a decade of plaice archival tagging

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Otolith oxygen isotope ratios are increasingly used to discriminate marine stocks, and applied as a proxy for temperature in current and past oceans, assuming that variations in otolith d¹⁸O values reflect differences in temperature history. To investigate how closely otolith d¹⁸O values reflect the environmental experience of wild fish, we examined the otoliths and corresponding archival tagging dataset (>13,000 geolocations and in situ temperature records from ~200 free-ranging fish) gathered from a decade-long study of plaice spatial dynamics and population structure in the North Sea. By focusing on otoliths from fish with known annual migrations over three consecutive years and corresponding, location-inferred high-resolution model temperature and salinity data, we compared predicted and measured annual otolith d¹⁸O values for three offshore sub-stocks with discrete annual distributions and temperature or salinity ranges. For individuals recaptured after >6

months at liberty, we also compared monthly $d^{18}\text{O}$ values accreted in the otolith during tag recording time with concomitant $d^{18}\text{O}$ predictions based on the tag temperature records. Both predicted (from temperature and salinity data using the isotope fractionation equation for inorganic aragonite deposition) and measured annual $d^{18}\text{O}$ values demonstrated >96% correct sub-stock prediction. Measured and predicted annual $d^{18}\text{O}$ values however, although largely harmonious, did not fully match, even when inter-stock differences in seasonal otolith growth were taken into account. Moreover, small but consistent offsets were observed between individual high-resolution otolith $d^{18}\text{O}$ values measured during tag recording time and corresponding $d^{18}\text{O}$ predictions. Possibly resulting from variation in physiological response to temperature among stocks, these results provide a compelling indication of small yet consistent vital effects during oxygen fractionation between ambient water and the mature fish otoliths. A fuller understanding of the mechanisms underpinning otolith $d^{18}\text{O}$ signatures is therefore desirable in the interpretation of otolith $d^{18}\text{O}$ data for stock discrimination or temperature reconstruction.

Abstract reference: IIIA_Huijbers_05

Stable isotopes in otoliths: what have we learned so far?

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Stable isotopes in otoliths are increasingly used to understand life history and movement patterns of fish. Due to the variability of isotopic values found in estuarine and marine ecosystems, and the inert nature of otoliths that keep a life-long record of the environment of a fish, isotopic analysis of otoliths can be used for a wide variety of purposes. We reviewed the current understanding of how measurements of carbon, oxygen and nitrogen stable isotopes in otoliths are used. Here, we specifically discuss advances in stock discrimination, and quantification of movement and contribution of juvenile habitats to adult populations. Classification of fish stocks yielded significantly higher success rates when carbon and oxygen isotopes were combined with elemental markers. The limited number of studies that successfully used isotopes to track actual fish movement showed unequal contributions from different juvenile habitats to adult stocks, which is crucial information for the sustainable management of fisheries. Based on our findings, we outline guidelines for successfully using stable isotope studies in different geographical regions. Overall, our analysis provides an imperative overview of the progress in this field, and directions for future research.

Abstract reference: IIIA_Hunter_06

Time and plaice: decadal changes in North Sea plaice *Pleuronectes platessa* L. observed through historical otolith and other legacy datasets

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Since the 1902 establishment of a small research station in Lowestoft to study the North Sea plaice fishery, The Centre for Environment, Fisheries and Aquaculture Science (Cefas) has advised the UK government on sustainable exploitation of marine living resources. From inception, Cefas have collected fisheries data and otoliths for age determination, both from market sampling and from surveys, while mark and recapture experiments were conducted to provide information about the movements and growth rates of fish, stock structure and mixing, and how these might affect fishery management. Although incomplete, these data represent a significant component of Cefas historical data archive, which in recent years has received high priority for cataloguing and digitising, and for developing permanent data storage solutions to better understand the past of our marine environment. To illustrate the value of our time-series, we use plaice as a model, showing a 113-year time-series of plaice length frequency from 1902 until present. We aim to analyze marked decadal changes in fish size with reference to what the fish were eating (from the DAPSTOM fish stomach content records database), and to their growth and survival through otolith analysis and age readings, and to examine whether these changes were systematic across all sub-stocks. Ultimately, we aim to discriminate whether observed changes relate to changes in bottom fauna (e.g. disappearance of bivalve beds with onset of extensive beam trawling), climate change, changes in fishing pressure (including times of reduced pressure during wars, reduction in current EU fleet capacity), and eutrophication. Our results provide a powerful illustration of the relevance of legacy data in strengthening the advice provided towards current and ongoing marine policy questions.

Abstract reference: IIIA_Mapp_07

Operational viability of stock-separation using shape indices derived from the otolith morphometric outline. An example using sprat and herring.

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We present a study concerning the viability of stock-separation of two highly mobile Clupeids (sprat *Sprattus sprattus* and herring *Clupea harengus*) using otolith morphometrics. Our analysis focuses on three stock discrimination problems with the aim of reassigning individuals to their source populations using a series of experiments undertaken using a popular suite of machine learning software known as the Waikato Environment for Knowledge Analysis. Within this framework 14

feature sets derived using 3 static transformations that encode combinations of size and shape were explored. By applying 9 state-of-the-art classification algorithms and using leave-one-out cross validation, we assessed how well each of the systems generalise to an independent data set. We performed 1260 cross-validated experiments on the data set, partitioned by age and using ground truth data provided by expert readers. To assess the saliency of size and shape features within each age partition, half of the feature sets include size indices whilst the remainder encode only shape. We found that for juvenile fish, feature sets that encode only shape perform well, but those that retain size indices hold more potential for classification. As specimens increase in age, discrimination based on morphometric features becomes ineffective; however as fish approach maturity transforms that encode shape appear to be slightly more robust to aging than those based on size. Using significance tests we show that for juveniles, our results with respect to transforms are significant but those with respect to classification algorithms are not. This study suggests that current operational methods of stock discrimination of Clupeids based on measurements of early incremental growth are likely to be effective, with little benefit in supplementing this information with shape indices derived from morphological outlines..

Parallel Session IIIB (Populations)

Abstract reference: IIIB_Marengo_01

Combined use of otolith shape, parasites and genetic markers for stock identification of the common dentex (*Dentex dentex*) around Corsica Island (NW Mediterranean)

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The common dentex *Dentex dentex* (Linnaeus, 1758) is an iconic marine coastal fish in the Mediterranean Sea. As a high trophic level predator, it holds a key position in the coastal marine food webs. It is highly valued by both commercial and recreational fisheries. *D. dentex* is now classified by the International Union for the Conservation of Nature (IUCN) as “vulnerable” in the Red List of Threatened Species in the Mediterranean Sea. Despite its economic and ecological importance, data on the stock structure of this species are still very limited. Information on stock identity and spatial structure are necessary for understanding fish population dynamics and enable reliable resource assessment for fisheries management. Consequently, a holistic approach is probably the most relevant way to define stock structure and favor the sustainable spatial management of an exploited species. The aim of this study was to examine the stock structure of the common dentex, using a combination of markers: otolith shape analysis (Elliptic Fourier descriptors, geometric morphometrics), parasites communities from the digestive tract and genetic markers (microsatellite loci). A sample of 103 specimens of *D. dentex* (46-91 cm total length) was collected in four locations around Corsica, in two consecutive years (2012 and 2013). The information produced by otolith shape analysis, parasites communities establishment and genotypic marker are combined in order to discriminate geographic ecological entities of *D. dentex* around Corsica. This study provides new insights into the population structure of common dentex at the spatial scale of the Corsica Island.

Abstract reference: IIIB_Hüssy_02

The Baltic cod: A case study for testing stock discrimination based on otolith shape analysis in a mixed stock fishery

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In the Baltic Sea two genetically distinct cod stocks are found, the “Eastern Baltic cod” and “Western Baltic cod”. In this study we evaluated the applicability of otolith shape analysis for classification of individuals caught in the fishery within the presumed mixing area, using genetic validation of the

otolith based assignments. The otoliths of eastern Baltic cod were generally wider in the dorso-ventral direction in relation to the anterior-posterior length than those of western Baltic cod. These differences were captured by otolith area and 7 – 10 Elliptic Fourier Descriptors of primarily low number harmonics. The genetic validation of otolith shape based classification of samples from the mixed-stock area revealed that the classification success using a baseline of spawning individuals was considerably lower than when a baseline of genotyped individuals including immature individuals also, was used. Stock mixing percentages using the latter documented that the “Eastern” cod stock has been present in the neighbouring management unit of the “Western” cod for many years. However, since 2005, their abundance has increased from 20% to >60% with a clear increasing age-related trend in presence. The observed immigration is not related to spawning migrations, as no seasonal effects on stock mixing were found.

Abstract reference: IIB_Brophy_03

Otolith shape variation in blue fin tuna from different regions of the North Atlantic: a possible marker of stock origin

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Two stocks of bluefin tuna (*Thunnus thynnus*) inhabit the North Atlantic; the western stock spawns in the Gulf of Mexico while the eastern stock spawns in the Mediterranean Sea. There is also evidence of finer scale structuring within the Mediterranean Sea. The species is highly migratory and trans-Atlantic movements are known to occur, resulting in mixing of the western and eastern stocks in the central North Atlantic. However, stock structure is maintained by high rates of natal homing. With both stocks severely depleted by over-fishing, a better understanding of population connectivity and the stock composition of mixed aggregations is needed to improve their management. This study aims to characterise Atlantic bluefin tuna from the western and eastern stocks based on otolith shape descriptors. Geographic variation in otolith shape within the Mediterranean which may indicate finer scale stock structure is also examined. Images of adult bluefin tuna otoliths collected from five areas (western, central and eastern Mediterranean; northeast Atlantic and northwest Atlantic) over three years (2011, 2012, 2013) were analysed using

elliptical fourier analysis. Regional differences in otolith shape were investigated using cluster analysis, while accounting for age and length related variability. The potential for using otolith shape measurements to complement information from otolith chemistry and genetics and to determine stock composition of mixed aggregations of bluefin tuna is discussed.

Abstract reference: IIIB_Fowler_04

Analysis of phenotypic characteristics of otoliths - resolving stock structure issues for snapper in South Australia.

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Snapper (*Chrysophrys auratus*) is a large, long-lived, demersal fish species with a broad geographic distribution through the Indo-Pacific region. It is one of the most significant coastal fishery species of Australasia, due to its broad distribution and high volume of catches. In Australia, snapper fisheries are managed at the jurisdictional level of States. The State of South Australia has recently dominated the Australian snapper catches. This reflected a period of dramatic change during which catches increased to record levels associated with a significant change in the spatial structure of the fishery. Whilst catches from traditional fishery regions of South Australia crashed, in several other regions they increased dramatically to unprecedented levels. The demographic processes behind these dramatic changes in biomass remain poorly understood. Fundamentally, there is considerable uncertainty about the stock structure throughout this geographic region, due to poor understanding of large-scale adult movement patterns. The analysis of otoliths provides a means of resolving such issues because these structures manifest physical and chemical differences based on spatial separation of fish, and because such differences are retained by the otolith that are not re-worked subsequent to deposition. In this study, sagittae, available from snapper from a long-term, market-sampling program, were selected from four strong year classes (1991, 1997, 2001, 2004) and six regions that cross a coastal distance that exceeds 1,000 km. Transverse sections were prepared from which numerous variables were measured, primarily along the long axis from the centre to the dorsal edge, including: length, age, increment widths, opacity and trace element concentrations. The analysis of these datasets will be interpreted in terms of where fish originated as juveniles, the likelihood of significant inter-regional movement throughout fishes' lives, and the possibility that any such movement has changed over time.

Abstract reference: IIIB_Stötera_05

Use of otolith quality flags to assess distributional dynamics in Baltic cod stock

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In the Baltic Sea, cod spawn in several basins that are separated by shallow sills. However, biological and management units of the two cod stocks do not match with the spawning grounds, causing constant concerns about mixing dynamics between ICES-Subdivisions (SD). Recently, Eastern Baltic cod displayed signs of recovery suggesting a spill over into the west. Basin-specific hydrographic conditions are reflected in optical differences and difficulties in age determination of Baltic cod otoliths, which may assist in evaluating the dynamics of mixing. We used 83 000 sliced and quality-flagged cod otoliths (readability categories: legible, uncertain or non-readable), collected from research surveys and commercial samplings of 2007 - 2013 covering ICES-Subdivisions SD21-29 to test the hypothesis of stable proportions of readability categories in time and space. The overall distribution of quality flags showed that the Darß and Drogden sill consistently separated large proportions of legible otoliths in the west (SD21-23) from large proportions of uncertain and non-readable otoliths in the east (SD25-29). SD24 clearly was a mixing area, however, strongly resembling SD25 and not SD22 or SD23. The relative distribution of quality flags only showed minor and inconsistent changes without significant trends in a multinomial logistic regression (grouped by all fishes, sex or length class), irrespective of the spatial (SD, rectangle, quarter-rectangle) and temporal scale (year, quarter). Instead, stable distribution patterns with about 3-5% of non-readable otoliths in SD22 and about 7-10% of legible otoliths in SD25 suggested stable mixing dynamics over time since 2007. The lack of increased proportions of unreadable otoliths in SD24 did not support a spill over from the east. However, the large proportion of uncertain otoliths in SD24 may mask the detection of mixing trends. The relationship between life history pattern of individual cod and quality flag category of their otolith are not yet understood.

Abstract reference: IIIB_Warren-Myers_06

Stable isotope otolith fingerprint signatures: a mass marking technique for farmed Atlantic salmon *Salmo salar*

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Understanding the ecological and genetic effects of fish escapees from aquaculture facilities on wild fish stocks requires the ability to trace escapees. This is currently difficult due to the lack of a cost effective, mass marking technique that differentiates wild from farmed fish and traces escaped fish back to the farm of origin. We investigated three stable isotope otolith fingerprint mark delivery techniques for mass marking farmed Atlantic salmon (*Salmo salar*) to determine if they could

achieve unambiguous marks with 100% accuracy. Otolith fingerprint signatures created with barium, strontium and magnesium isotopes were delivered via: 1) Maternal transfer: where an isotope marker is injected into broodstock, and thus in turn passed on to the offspring; 2) Egg immersion: where fertilised eggs are left to swell in a solution containing an isotope marker; and 3) Vaccination: where the isotope marker is combined with a vaccine and injected into fish. Analyses of otolith signatures were carried out on a Varian Inductively Coupled Plasma Mass Spectrometer (ICP-MS) fitted with a HelEx (Laurin Technic and the Australian National University) laser ablation (LA) system constructed around a Compex 110 (Lambda Physik) excimer laser. 100% marking success was achieved with all three techniques at specific marker concentrations and all techniques are capable of creating marks that cannot be confused with marks in wild fish. Mark success was dependent on type, concentration, and combination of isotope markers used. In addition, no measurable side effects of stable isotope marking on fish growth, condition, or mortality were observed. The most promising technique, vaccination, can produce multiple fingerprint signatures for as little as 0.1 US cents per fish. We conclude that mass marking farmed Atlantic salmon with stable isotope otolith fingerprint signatures is a viable method to identify and trace farmed escapees.

Abstract reference: IIB_Lauchlan_07

Stable nitrogen isotopes in otoliths discriminate juvenile fish stocks in estuaries affected by anthropogenic impacts

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Coastal estuarine habitats are regarded as important nurseries for fishes, but the first step to assessing the contribution of inshore areas to adult populations offshore is to distinguish different juvenile stocks. In many estuaries, natural habitats and fish populations are influenced by urbanisation and industrialisation of the coastline. Stable nitrogen isotopes ($\delta^{15}\text{N}$) can be a good indicator of anthropogenic influences, but are rarely measured in otoliths because they occur in such small amounts. We tested if $\delta^{15}\text{N}$ of fish otoliths could provide a useful tracer for stock discrimination of fish in estuaries affected by anthropogenic impacts. Juvenile Moses perch (*Lutjanus russelli*) were collected from five sites, which differed in distances to urban development and salinity, in southern Queensland, Australia. Otolith $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ isotope values were measured by combustion. Both $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ values differed among sites, and a combination of both isotopes resulted in a high classification success (88%). Nitrogen isotope values were significantly enriched in otoliths from fish collected at inshore sites heavily influenced by anthropogenic development. Our study demonstrates that, despite only small amounts of nitrogen present within otoliths, $\delta^{15}\text{N}$ can successfully be analysed and used as a tool to discriminate among different areas of putative juvenile habitat.

Abstract reference: IIIB_Patterson_08

Population Mixing between U.S. King Mackerel Stocks Estimated with Otolith Chemical Signatures

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King mackerel, *Scomberomorus cavalla*, is a coastal migratory pelagic species with distinct migratory groups in U.S. waters. Atlantic Ocean (Atlantic) and eastern Gulf of Mexico (eGOM) fish migrate to south Florida in late fall where various fisheries target the mixed populations throughout winter. We employed natural tags based on otolith chemical signatures (Ba:Ca, Li:Ca, Mg:Mn, Mn:Ca, and Sr:Ca) to distinguish Atlantic from eGOM fish from 2004-2008, and then applied the signatures to estimate the contribution of the Atlantic versus eGOM population to winter mixed-stock fisheries among three south Florida regions. Otolith chemical signatures analyzed from fish sampled in the Atlantic and eGOM during summer were significantly different between populations and among sampling years (MANOVA, p90%) fish sampled off southeast Florida were estimated to be Atlantic fish. A second trend that existed was that the estimated contribution of eGOM fish to winter south Florida fisheries increased across the time series. This increase was consistent with otolith shape analysis results and was likely due to increasing spawning stock biomass in the eGOM population as it recovered from overfishing during the 2000s. Results of this work have important implications for the management of this highly valued fishery species and have been incorporated into statistical catch at age stock assessment models in which mixing between Atlantic and eGOM populations is modeled.

Abstract reference: IIIB_Morais_09

European flounder life history plasticity: paranormal activity or the helpful insights of otolith microchemistry?

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Fish life history plasticity research is among the least studied topics by ichthyologists, but is benefiting from advances in otolith microchemistry, mostly in diadromous species. The European flounder is among those few studied species that shows various alternative tactics to catadromy. Otolith microchemistry data from flounders collected in four estuaries (Lima and Minho in Portugal, Gironde and Seine in France), plus two adjacent coastal areas, indicated that there were three main life history patterns: A) inland spawning, inland restricted development, B) inland spawning, inland/coastal development, C) coastal spawning, coastal/inland development. Please note that

inland stands for an estuary, either their brackish or freshwater sections, or to a river. The diversity and frequency of each life history pattern varied between ecosystems, and it was not possible to observe any clinal trend. There has been some controversy around this finding, but otolith microchemistry data points in the same direction as anecdotal evidence- catadromy is just one of the strategies used by the European flounder. We suggest that the coexistence of alternative life history strategies might result from a successful evolutionary process to maximize individual fitness. Thus, fish life history plasticity is a trait that enable species to persist in varied and unpredictable environments, which could be highly beneficial in the current context of climate change. Finally, a growing number of otolith microchemistry studies suggest that fish life history plasticity might be more common than expected, so scientists should be more receptive to the possibility of its occurrence, either when assessing a species life history or when proposing fishing policies.

Parallel Session IIIC (Dispersal)

Abstract reference: IIIC_Kawakami_01

Ecological changes in and recovery of the Ayu population following the tsunami generated by the 2011 Tohoku earthquake

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The tsunami generated by the 2011 Tohoku earthquake devastated the coastal environment of the Tohoku region, destroying coastline habitats and altering the species composition and biomass of benthos and fish assemblages. These disturbances are assumed to have affected the ecology of the Ayu, *Plecoglossus altivelis altivelis*, which has a 1-year amphidromous life history. To reveal the ecological changes in and recovery of the Ayu population, the species' migration history was investigated by analyzing otolith increments and Sr:Ca ratio of fish in rivers located in the Sanriku region (the Unosumai and Sakari rivers) during 2011–2013. The results obtained were compared with data collected before the tsunami (2010). In 2011, all fishes collected in June were late-hatched (after October), in contrast to 2010, in which early-hatched (during September) fishes accounted for 52–62% of the population. In both rivers, the mean hatching date had shifted to a significantly later date (Unosumai River: September 28 in 2010 and October 16 in 2011; Sakari River: September 30 in 2010 and October 20 in 2011). In addition, in 2011, the fishes ascended the rivers at a significantly younger age than in 2010, despite similar ascending dates (early or mid-May). Therefore, the tsunami probably induced a high mortality of early-hatched juveniles distributed around the estuaries just before they entered the rivers. During the following 2 years, delays in the hatching date and spawning season were observed in the Unosumai River until 2012, although in both rivers in 2013, hatching and spawning had recovered to times similar to those before the tsunami. These results suggest that recovery of the ayu population may have differed depending on the environment. To clarify the recovery of ayu from the impact of the tsunami, detailed information on survival and growth in the ocean together with population genetic studies are required.

Abstract reference: IIIC_Egan_02

Out of sight, out of mind: what can otolith microstructure tell us about the elusive marine dispersive phase of the New Zealand whitebait *Galaxias maculatus*?

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Galaxias maculatus is one of five diadromous Galaxiids native to New Zealand. The translucent juveniles (whitebait) comprise >90% of a fishery that has important cultural, recreational and commercial values. Several intrinsic and extrinsic factors make an ecosystem-based management

approach to the conservation and sustainable harvest of this species challenging. These factors include a bipartite life cycle with uncertain larval development and dispersal, panmixia, the multispecies nature of the whitebait fishery, as well as cultural, political and socio-economic concerns. Our wider programme has addressed spawning habitat restoration of degraded waterways to increase egg production, thereby supplementing the fishery, but here we examine the challenging problem of the marine larval phase. Since Panella's (1971) discovery of daily ring deposition in otoliths, the task of understanding the marine larval phase of bipartite species has become more achievable. The aim of this research is to use otolith microstructure characteristics to explore the 'black box' of larval development and identify components that are spatially or temporally discrete during different phases of *G. maculatus* life cycle. Otoliths were collected bi-monthly from four spatially discrete fisheries in New Zealand throughout the 2013 fishing season. Age and size at migration, pelagic larval duration, growth rate at sea and other characteristics were derived. Daily growth rings confirm larval development periods and variation in microstructure characteristics within and among regions is evident. In particular, several recruitment events into rivers occur throughout the year offering the opportunity to examine temporal microstructure properties. The information obtained from these calcified structures provides biologically meaningful data that can be used to inform scientists and policy makers, and help in better management of New Zealand's iconic whitebait fishery.

Abstract reference: IIC_Herrera-Reveles_03

Pelagic larval duration and growth of early life stages of a coral reef fish: variations in an upwelling South Caribbean area

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The goal of this research was to evaluate the variation in pelagic larval duration (PLD) and growth rates during early life stages of *Abudefduf saxatilis* (Pomacentridae) in a Venezuelan upwelling area, South Caribbean. We analyzed the sagittae otolith microstructure in monthly collected recruits during a year following a nested design at three spatial scales. We used the pattern of ring daily growth on the otoliths to determine the birthdates of recruits, and we measured the pelagic stage duration and growth rate using the variations in the otolith's rings width. PLD was between 20 and 31 days. The shorter periods were registered during upwelling events (January – March 2011) (PERMANOVA, $p < 0.05$). These results suggest that survivors born during the period of low environmental stability (upwelling months) would be those that have shortest PLD and those settled faster from the plankton to the benthic habitats. Relationships between body size, age and otolith diameter were significantly and positively correlated, and fitted to a linear regression model suggests proportionality between otolith growth and fish somatic growth. The values of recent growth rates ranged from 0.226 to 0.384 mm day⁻¹, and differed between sites and months (ANCOVA, $p < 0.05$). The lowest values take place at low temperatures (22.7-24.39 °C) and high upwelling indexes (1.64-1.88 m³ seg⁻¹). The values of back-calculated growth rate of larval stages oscillated between 0.072 - 0.188 mm day⁻¹, and differences were not found, whilst back-calculated growth rate at post-larval stages have a similar temporal patterns to recent growth rate (0.269 and 0.606 mm day⁻¹) (PERMANOVA, $p < 0.05$). These variations suggest that higher growth rate occurs during the higher environmental stability period, when wind speed is low (2.2-2.6 m seg⁻¹), so that turbulence mixed water that could disturb growth rates during post-larval stages are not generated.

Abstract reference: IIIC_Freshwater_04

Interpopulation and individual variation in dispersal characteristics of juvenile Sockeye Salmon

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On the west coast of North America, juvenile Sockeye Salmon (*Oncorhynchus nerka*) generally appear to undertake a rapid northward migration after marine entry. Variation in dispersal among and within populations has been increasingly documented and may play an important role in regulating stock-specific survival rates. Unfortunately, assessing differences in ecological characteristics that may drive variation in dispersal characteristics is difficult using catch data alone. In this study, we used otolith microstructure to reconstruct the phenology and growth history of four populations of juvenile Sockeye Salmon captured from southern British Columbia to the Alaskan border in 2007 and 2008. We tested the hypotheses that coastal dispersal is affected by variation in 1) migratory rate, 2) ocean entry size, 3) ocean entry phenology, and 4) growth at sea. We observed that within a population, individuals captured further along the migration corridor were larger at capture, dispersed more rapidly, and had entered the marine environment at a larger size than the rest of their cohort. Conversely, ocean entry date did not consistently differ between capture locations. We also observed that while weekly growth rates consistently increased during ocean residency, they did not differ between sampling regions. While these broad trends were robust across years and populations, the strength of the relationship often varied inter-annually and with an individual's region of origin. Therefore dispersal in juvenile Sockeye Salmon appears to be more strongly influenced by size at ocean entry and inherited, stock-specific traits than entry phenology or local differences in growth conditions experienced at sea. The divergent dispersal strategies we observed may act to reduce density dependent competition in near-shore environments and buffer populations from mortality incurred during migration.

Abstract reference: IIIC_Shima_05

Are larval reef fish travelling in packs? Using otoliths to evaluate evidence for shared dispersal histories

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The larval stage of reef fishes is disproportionately important for population dynamics and evolutionary processes, yet it remains poorly understood. In part this is because larval fish are small and difficult to track in a large and turbulent ocean. We use information recorded within the otoliths of a small temperate reef fish, the common triplefin (*Forsterygion lapillum*) to reconstruct larval developmental histories to address a series of questions with important implications for metapopulation dynamics and evolutionary processes. We used light traps to collect larvae at

discrete sites and times around Kapiti Island, New Zealand. For a sample of individual larvae from each site and date, we quantified otolith microstructure, reconstructed daily growth histories, and expressed these as 'hindcasts', i.e., ordered from date of capture (outer edge of otolith) to date of hatch (edge of otolith core). We quantified otolith microchemistry along this same axis of measurement (using LA-ICPMS) and used hindcasted growth history to estimate a daily record of trace element concentrations for each fish, from point of capture back in time to point of hatch. Using this data set, we then asked: (1) How many discrete "dispersal cohorts" supply a given site on a given night?; (2) Is there any evidence that dispersal cohorts form during pelagic development or at the natal site?; (3) Do patterns vary among sites and/or through time? Our results suggest that the number of dispersal cohorts varies from 2 to 8 among sites and dates, and that some cohorts appear to form early and persist through much of the pelagic larval duration (suggesting a common dispersal history), while others form shortly before capture. We discuss the possible causes and important implications of these results for population dynamics and evolutionary processes.

Abstract reference: IIC_Martin_06

Dispersal capacities of Allis Shad (*Alosa alosa*) under global change: insights of innovative otolith microchemistry analysis

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We aimed at assessing the dispersal capacity of Allis shad, an anadromous clupeid in decline throughout its distribution range. Colonization of new environments by shad is achieved via straying behavior, framed as failure to home, a fundamental life-history trait present in various proportions in anadromous fish populations. Geochemical signatures recorded in otolith were chosen to identify natal origin of 410 spawners caught at spawning grounds along the Atlantic coast from northern France to Portugal. However, as juveniles display a short freshwater residence time, a specific laser ablation strategy was required to target precisely the two weeks after hatching on the otolith. A novel UV high-repetition-rate femtosecond laser ablation device was deployed, permitting ablation of 60 µm thick coronas around otolith primordium, while avoiding any maternally-derived material accreted at the core. Otolith and water samples were analyzed for Sr:Ca, Ba:Ca, Li:Ca and ⁸⁷Sr:⁸⁶Sr ratios using a High Resolution ICPMS and a Multi-collector ICPMS. The allocation of natal origin was obtained from different sources of information (water chemistry, signatures in otolith of juveniles and spawners) within a Bayesian hierarchical mixture model. The chief advantage of this method over frequentist approaches was to exploit the relationship between water and otolith signatures, providing otolith chemistry to be predicted, with a mean classification accuracy of 86%, even for rivers where juveniles were unavailable. Results showed that 35% of adults displayed a straying behavior, mostly originating from neighbouring rivers. The proportion of strayers differed between source populations. Among the seventeen sampled locations, only three acted as a source population supplying adults to adjacent or distant sites. The southernmost population displayed the highest dispersal potential and 18 individuals from this population strayed up to 1800km to the

northernmost French river. Neither total length at maturity nor sex determined the spatial scale at which straying occurred.

Abstract reference: IIC_Di Franco_07

Investigating early stages dispersal using otolith chemistry: surprising relevance of the post-settlement phase in a temperate coastal fish.

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The concept of dispersal acquired a large significance from a management perspective, particularly for the design of networks of marine protected areas (MPAs). Generally, dispersal of demersal fishes is equated with propagule (*i.e.* eggs and larvae) dispersal, neglecting the potential of dispersal at successive stages. Using otolith chemistry analyses we estimated dispersal patterns at propagule and juvenile/sub-adult (*i.e.* between settlement and recruitment) stages of a Mediterranean coastal fish, the two-banded seabream *Diplodus vulgaris* (ecologically important predator and target species for fishing). In SW Adriatic Sea (Italy, Mediterranean Sea) we collected post-settlers (n=157) and recruits (n=164) in 14 sampling sites spread along about 180 km of coastline. Basing on elemental signatures in otolith cores we identified groups of post-settlers corresponding to different natal sources and we estimated larval dispersal on the basis of the distance among sampling sites replenished by a single natal source. We obtained information about juvenile/sub-adult dispersal by evaluating "site fidelity" and/or the distance travelled between settlement and recruitment sites. The elemental composition of the otolith portion formed just after settlement of post-settlers was used to generate a reference set of site-specific chemical fingerprints. Then, a similar analysis (*i.e.* on the same portion of the otolith) was done on recruits. Post-settlement dispersal was inferred assigning recruits to their settlement sites. We detected three major natal sources replenishing with propagules the entire study area, suggesting that propagule dispersal could take place across at least 90 km. We also recorded an extensive dispersal, up to 165 km, at juvenile/sub-adult stage. Here, we provided information about effective dispersal at propagule and juvenile/subadult stages for a coastal fish, pointing out a non-negligible role of juvenile/subadult life stages in connecting populations through extensive dispersal. This represents a piece of information crucial to build up ecologically sounded management strategies (e.g. effective MPAs' networks).

Abstract reference: IIIC_Tanner_08

Assessing fish population connectivity: Combining otolith geochemistry and biophysical models

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Understanding connectivity in fish populations and quantifying dispersal and movement of fishes is a challenging task. Depending on the life stage where movement is measured, different approaches have yielded remarkable results. Coupled biophysical models have significantly contributed to advance our understanding of larval dispersal in marine fish. Otolith geochemistry has developed into an outstanding tool to determine connectivity, population structure and dynamics. Nonetheless, the use of multiple techniques that provide information over different scales and life history stages might provide the best inferences on population connectivity and structure particularly, in species that occupy spatially segregated habitats throughout their life history. Here, data on connectivity between different life stages of common sole (*Solea solea*) along the Portuguese coast are presented. Adults of this species live and spawn in coastal areas while juveniles use estuaries as nursery areas. Larval dispersal towards estuarine plumes is assessed using biophysical modelling while juvenile nursery contributions from estuaries to the coastal adult stock are estimated through otolith geochemistry. The combined use of the two techniques provides a more comprehensive understanding of the rate and extent of connectivity over the different life history stages. In addition, the input from each technique towards connectivity assessments is discussed. Ultimately, such information may be essential for the development of effective conservation strategies and integrated fisheries management plans.

Abstract reference: IIIC_Barnett_09

Estimating potential nursery sources for red snapper, *Lutjanus campechanus*, populations in Atlantic Ocean waters of the United States from North Carolina to Florida

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Red snapper, *Lutjanus campechanus*, are managed as a single stock in Atlantic Ocean waters of the United States from North Carolina to southern Florida, but little information exists to infer population structure. We examined otolith chemical signatures ($\delta^{13}\text{C}$, $\delta^{18}\text{O}$, Ba:Ca, Mg:Ca, Mn:Ca, and Sr:Ca) in the core of adult (ages 3-6 y) otoliths sampled in 2010 from two clusters of samples,

one south of 30N and one north of 31N, to examine if chemical signatures differed between regions or ages. There were significant differences between regions (MANOVA; $p < 0.001$) but not age groups (MANOVA; $p = 0.104$), although only ages 4 and 5 co-occurred in samples from both regions. Constituents contributing to the significant difference between regions were $\delta^{13}\text{C}$ (ANOVA; $p < 0.001$), Ba:Ca (ANOVA; $p < 0.001$), and Sr:Ca (ANOVA; $p = 0.002$). These results indicate distinct region-specific signatures exist, thus suggesting differences in nursery sources for each cluster of samples. However, otolith chemical signatures remained more or less consistent among age classes for southern fish, while among northern samples the distribution of signatures broadened and became less distinguishable from southern samples with increasing fish age. This pattern may suggest increased post-settlement movement and population mixing with age, which has been reported for this species in the Gulf of Mexico. Ongoing Markov Chain Monte Carlo analysis is being employed to estimate the number of potential nurseries contributing to adult populations in each southeastern United States Atlantic Ocean region, which is aimed at statistically deciphering observed patterns in otolith chemical signatures.

Abstract reference: IIIC_Reis-Santos_05

Connectivity and fish population structure: perspectives from otolith geochemistry and genetic markers

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Connectivity and population structure are influenced by physical, biological and ecological processes and interactions that impact over a range of temporal scales. Therefore the best inference on population connectivity may be achieved by using multiple and potentially complementary techniques that integrate over different spatio-temporal scales. In the present study, we examined natal origin, movement and population structure of European flounder *Platichthys flesus* and Senegalese sole *Solea senegalensis* by combining otolith geochemical composition and microsatellite DNA markers of fish collected from multiple areas along the northeast Atlantic Ocean and Mediterranean Sea. The integrated assessment of otolith and genotypic markers enhanced the present connectivity assessment. Overall, the two natural markers were complementary; otolith geochemical data provided information on ecological time frames and finer spatial scales, while microsatellite DNA markers highlighted gene flow over broader time scales. The added value of using otolith geochemistry to complement the assessment of early life stage dispersal in marine populations with high gene flow is discussed, as well as the advantages and potential pitfalls of objectively integrating data from these different natural markers. Ultimately, unravelling connectivity and population structure via multiple methodological approaches are increasingly important to identify management requirements for these commercially important species.

OtoChuki Session (IIID, Tuesday)

Abstract reference: IIID_Fraile_01

Movements of the North Atlantic albacore (*Thunnus alalunga*) and bluefin tuna (*Thunnus thynnus*) revealed by otolith $\delta^{18}\text{O}$, $\delta^{13}\text{C}$, Sr:Ca, Mg:Ca, Mn:Ca and Ba:Ca chronologies

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Trace elements and stable isotopes in otoliths of North Atlantic albacore tuna (*Thunnus alalunga*), collected from the different feeding grounds in the eastern North Atlantic were measured and compared among sampling locations and life history stages. Elemental and isotopic fingerprints measured in core and edge zones were used to determine if albacore tuna captured at different locations (inshore waters of the Bay of Biscay vs. Atlantic offshore waters including southern Celtic Sea) have the same nursery origin and migratory patterns. Otolith edge geochemical signatures proved to be a valuable tool for classifying individuals to their capture locations. Multivariate analysis of the trace element and stable isotope values of the otolith edge reflected a clear differentiation between sampled locations. Among the element analysed, Sr concentration appeared to accurately reflect ambient physicochemical conditions, and served as a good proxy to differentiate capture locations. $\delta^{18}\text{O}$, $\delta^{13}\text{C}$, Sr:Ca, Mg:Ca and Ba:Ca during the early life history stages (0 to 6 month of life) were relatively invariant across the two capture locations, but Mn concentrations differed significantly among albacore tuna captured in coastal waters of the Bay of Biscay and in Atlantic offshore waters. Mn concentrations were higher in fishes captured in the Bay of Biscay, suggesting a higher growth rate during the early stages compared to fishes captured in offshore waters. Annual cycles of Sr:Ca ratios were visible along the otolith ablation transects reflecting an annual migratory pattern between water masses of differing salinities. Sr:Ca cycles were visible during the early life history stages for most of the juvenile albacore captured in the Bay of Biscay, coinciding with a high accumulation of Sr during the winter, whereas this periodicity was visible only in later stages in fish from offshore waters.

Abstract reference: IIID_Moore_02

Stock identification of *Sprattus sprattus* within the Celtic Sea Ecoregion using otolith shape and microstructure

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Sprat (*Sparattus sprattus*) is a species of immense ecological and economic importance. Its highly variable recruitment and consequent fluctuations in biomass present a challenge to the sustainable management of the sprat fisheries. In the Celtic Sea Ecoregion (ICES Divisions VIa, VIIa, VIIb, VIIg, VIIh, VIIj) fishing effort for sprat has increased due to an increased demand for fishmeal and the

reduction of quotas for other fisheries. However, the stock is not currently assessed due to data deficiencies and a poor understanding of the stock structure within the region. The objective of this study is to examine geographic variations in otolith shape and microstructure with a view to using these techniques for stock delineation. Both of these methods have been successfully used to elucidate stock structure in other species where a significant level of genetic separation is undetectable. Sprat were collected from two key fishing areas in the north and south of the region (ICES Div. VIa and VIIj) throughout the 2013 fishing season. Otolith outlines were extracted from digital images and described using Elliptical Fourier Descriptors. The widths of daily increments within the larval region of the otolith were used to determine if fish from the two areas had distinct larval growth histories. The results are discussed in relation to defining appropriate stock boundaries for sprat in the Celtic Sea Ecoregion as part of a sustainable management strategy.

Abstract reference: IIID_Rulifson_03

Residency and Habitat Use of Southern Flounder in North Carolina Coastal Watersheds

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Southern Flounder *Paralichthys lethostigma* is known to utilize coastal rivers as critical habitat but duration of the residency is unknown. Estuarine critical habitats of preferred soft-bottomed conditions are being threatened by dredging operations, shoreline development, mining operations, and bottom disturbing fishing gear. We examined Southern Flounder freshwater habitats use using multiple techniques: fatty acid profiles of muscle tissue, total mercury levels of the same tissues, and otolith chemistry. All three techniques confirmed that once entering and residing in coastal freshwaters, Southern Flounder remain until they are old enough to migrate to sea for spawning. Reduced growth and low abundance are several indicators that coastal rivers are not optimal habitat for Southern Flounder, but should be considered as important secondary habitats as estuarine conditions shift with climate change and anthropogenic influence.

Abstract reference: IIID_Khan_04

Discrimination of *Sperata seenghala* stocks inhabiting three rivers of the Gangetic river system using elemental fingerprints on otoliths

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The giant river catfish, *Sperata seenghala*, is one of the most important bagrid catfish captured in Indian subcontinent. The wild population of the fish has shown a declining trend over the years but there is paucity of published information on its population structure. However, a comprehensive knowledge on the stock structure is necessary for proper management and conservation of the

target fish species. Therefore, the present study was undertaken with a view to discriminate the stock(s) of *Sperata seenghala* population collected from Narora, Kanpur and Bhagalpur sites on the Ganga River, Firozabad site on the Yamuna River and Lucknow site on the Gomti River using elemental fingerprints on otoliths. Fifteen trace elements from whole sagittal otoliths were analysed using inductively coupled plasma atomic emission spectrometry. For each site, elemental concentrations were subjected to appropriate uni- and multivariate statistical tests. Strontium, barium, lithium, copper, iron, lead, zinc, manganese, nickel ($p < 0.001$) and magnesium ($p < 0.01$) differed significantly among locations, while no significant differences were noted for calcium, sodium and potassium ($p > 0.01$). Chromium and cadmium were not detected in the otoliths of the fish from Narora site on the river Ganga. Discriminant function analysis using cross-validation classification, assigned individuals to their site of sampling origin with a mean classification accuracy of 83.2%. The detected site-specific elemental differences in *S. seenghala* otoliths indicate a high level of site-fidelity in relation to their habitat areas. The target fish population from these sites can be regarded as heterogeneous stocks forming separate units for fisheries management in the selected rivers.

Abstract reference: IIID_Lewis_06

Distinguishing Blacktip Shark, *Carcharhinus limbatus*, Nursery Areas in the Northern Gulf of Mexico with Vertebral Chemical Signatures

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The life history traits of large coastal sharks make them particularly susceptible to overfishing. Interest in incorporating a habitat component into management plans has resulted in a focus on conserving shark nurseries. However, we currently lack estimates of connectivity between juvenile and adult populations making it difficult to evaluate the importance of nursery conservation to population stability and recovery. The analysis of trace metals incorporated into otoliths of bony fishes has emerged as a powerful tool for estimating connectivity between juvenile and adult populations. For coastal elasmobranchs, it may be possible to infer natal origin based on nursery-specific biogeochemical signatures in their vertebrae. To assess the efficacy of this approach, we collected neonate and young of the year blacktip sharks, *Carcharhinus limbatus*, ($n = 41$) from three regions (Florida, Alabama/Mississippi, and Texas) in fall 2012 and analyzed their vertebral centra with laser ablation-inductively coupled plasma-mass spectrometry (LA-ICP-MS). Three parallel ablation transects were assayed to construct trace metal maps and estimate region-specific biogeochemical signatures. Maps of trace metal concentrations indicate Mn:Ca peaked following birth. Both Sr:Ca and Ba:Ca also exhibited distinct shifts concurrent with birth. Biogeochemical signatures were significantly different among regions (Pillai's Trace = 0.857, $p < 0.001$), and linear discriminant function analysis yielded a mean regional classification accuracy of 72%. Texas had the lowest classification accuracy (55%); however, ablating greater vertebrae mass may increase the number of elements above detection limits, thus increasing discriminatory power. This is the focus on ongoing research, along with examining interannual variability in nursery signatures.

Abstract reference: IIID_Dobbs_07

Natal Origin of Pamlico Sound, North Carolina, Striped Bass Inferred from Otolith Chemistry

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Striped Bass *Morone saxatilis* populations of the Neuse and Tar-Pamlico watersheds were examined using LA-ICP-MS on adult otoliths to determine whether hatchery stocking was successful in these systems. Fortuitously, but unknowingly, the methodology used to rear Striped Bass in North Carolina produced a clear hatchery signal in the otolith. The signal was caused by extraordinarily high strontium concentrations in the underground aquifer water used to rear the fish to Phase I or Phase II status for stocking. Wild fish did not have this high strontium signal for the first year of life. Using our developed criteria based on the signal, we estimated that 88% of all sampled adult fish in both watersheds were of hatchery origin, suggesting that these two populations now support only a put-and-take fishery. Apparently there is natural recruitment failure by wild fish to the recruiting year classes, but the cause of this failure is undetermined.

Abstract reference: IIID_Şahin_08

Using Otolith Shape Analysis as a tool of stock discrimination of the anchovy subspecies in the Black Sea

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In the Black Sea, anchovy is a commercially important small pelagic fish source which represented by two different subspecies, hence, by two different stocks. They have different spawning and foraging areas and overlapping wintering ground. They have a tendency to be in the north and in summer time where the nutritious conditions are better. With winter cooling, they migrate downward towards the warmest southeastern corner of the Black Sea. The schooling behaviour makes them primarily target species of the Black Sea fishing fleet. Due to morphological similarities, it is not possible to report the catch by subspecies and this creates crucial problems in stock assessment. The discrimination methods such as genetics, parasitism rate, blood types are either not practical or very expensive. The otoliths, which are fundamental for the analytical stock assessment techniques, provide sufficient information about the stock units. In that respect, feasibility of the use of otoliths in the stock recognition of Black Sea anchovy has been tested in this study. With this aim, anchovies sampled during the July 2013 Black Sea ichthyoplankton survey, when the stocks move apart, have been analyzed using multivariate analyses. The approach was to start with as much morphometric variable determined on the otoliths as possible and reduce them to simply the discrimination. The shape parameters determined based on separating eastern and western summer anchovies were used to define the origin of the mixed overwintering anchovies sampled during the fishing seas in winter.

Abstract reference: IIID_Rohtla_09

Provenance and migration patterns of European whitefish *Coregonus lavaretus* (L.) s.l. in the Baltic Sea – combining otolith geochemistry and gill raker counts

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European whitefish (*Coregonus lavaretus* L.) is an ecologically, commercially and recreationally important fish species in the brackish Baltic Sea and in its freshwater basins. Three general life history types exist: (1) seawater residency, (2) anadromy and (3) freshwater residency. European whitefish is known to perform long spawning and feeding migrations that can transcend country borders. In addition to naturally reproducing populations, vast numbers of hatchery origin fish are stocked yearly in different countries surrounding the Baltic Sea. However, the proportion of hatchery origin fish in the catches is largely unknown. In this study we investigated provenance and migrations patterns of European whitefish using otolith Sr:Ca and ⁸⁷Sr:⁸⁶Sr values in combination with gill raker counts and published geochemical literature. In order to investigate the natal baseline values of freshwater origin fish, young-of-the-year (YOY) fish (n=55) were collected from four Finnish hatcheries and from one site close to the mouth of Pärnu River, Estonia. Thereafter, natal zones of otoliths from adult specimen (n=82) collected from six Estonian coastal sites were analysed to see if it is possible to distinguish: (1) general life history types (2) hatchery origin fish and (3) natural fish from Estonia and from other basins. Migration patterns were inferred from otolith core-to-edge Sr:Ca profiles. The results indicated that YOY fish from the Finnish hatcheries could be distinguished with high confidence (reclassification rate 100 %). YOY fish from Pärnu River were typical natural migrants/drifters with minimal freshwater signal. Almost 50 % of the adult fish caught from the Estonian coast were from hatcheries that are most likely located in Finland. These fish had resided in freshwater for prolonged period during early ontogeny, indicative of typical over-summer rearing in hatcheries and stocking in autumn. Individuals with low gill raker counts (indicative of local origin) never possessed a natal fingerprint produced by non-Estonian hatchery.

The validation of otolith traceability tools within a robust, reproducible and transferable forensic framework

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EU fisheries monitoring and enforcement increasingly requires legal evidence of provenance for individuals or batches of fish. The enhanced visibility of fish traceability tools is an important legacy of the FishPopTrace project (FP7-KBBE-2007-1) (<https://fishpoptrace.jrc.ec.europa.eu/>). The use of scientific results to provide legal evidence requires forensic validation of the techniques and analytical protocols, which must be developed beyond the level of a research methodology, up to the standard of a robust, accredited forensic assay. To bring otolith composition analysis (OTOMC) and otolith shape analysis (OTOMP) into consideration as traceability tools, we optimized standard operating procedures (SOPs) documented in a format suitable for intra- and inter-laboratory validation on the FishPopTrace target species: cod, hake, herring, and sole. The OTOMP validation SOPs emphasized required image and data quality, rather than exact duplication of equipment, to allow direct validation of results and fitness for purpose. Intra- and inter-laboratory precision achieved the required criteria of CV < 3% for 99% of the shape measurements. Circularity and perimeter were the most variable measurements, but were not significant factors for the statistical assignment of individuals to populations. The OTOMC validation exercise was based on repeated measurement of matched samples, prepared simultaneously from the “mirror face” of each otolith. Accuracy was determined by comparison with otolith reference materials (NIES-22, FEBS-1). Precision was determined by hierarchical comparison of measurements within otoliths, within operator at different sessions (intra-laboratory), and between operators/machines (inter-laboratory). The best precision for cod and hake was CV=2% for Ca, but was ~20% for Mg and Sr. CV%*s* were always higher in herring and sole, probably due to the smaller size of their otoliths. Precision was better for more abundant elements, and was usually better for line mode ablation at the otolith edge compared to spot ablation at the otolith core. In conclusion, the OTOMC criteria were more difficult to achieve, and species-specific suites of elements are likely to be required for validated traceability tools. In contrast, the otolith dimensions used for OTOMP are repeatable with good precision, and contribute to consistent assignments of individuals. Thus OTOMP could readily become a validated traceability tool.

Abstract reference: IIID_Gatti_13

Migration patterns and population structure of two small pelagic species from otolith microchemistry: European anchovy (*Engraulis encrasicolus*) and Sardine (*Sardina pilchardus*) of the Bay of Biscay

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Life history and connectivity between essential habitats for successive developmental stages are critical knowledge to better understand dynamics and distribution of marine fishes. These concepts encompass multiple processes at individual or population scale, eg. growth, feeding, mortality and movement. To deal with these issues a variety of tools can be implemented e.g. modeling, tagging, genetics, otolith shape or microchemistry analysis. Otolith microchemistry has widely been used to identify stock structure or migration patterns, using metal elements or stable isotopes. Indeed, there is a lack of understanding and knowledge on dispersal and migration patterns of fish. To identify potential migration patterns and « sub-population » of two small pelagic species, namely European anchovy (*Engraulis encrasicolus*) and Sardine (*Sardina pilchardus*), in the Bay of Biscay, otolith microchemistry has been implemented using a LA-ICPMS. As a first testing on these oceanic species, around 45 samples per species were taken from PELGAS pelagic survey in Spring and EVHOE bottom trawl survey in Autumn. In addition, some juvenile « white anchovies » (*Engraulis albidus*) were also sampled. Due to their expected residence in the Gironde river plume and its surroundings otolith element signatures would be typical of this coastal area for this genus. A set of metal elements was tracked: Sr, Ba, Li, Mn, Mg, Cu, Cd, Zn, Pb and Hg. Cd, Cu and Zn are typical markers of the Gironde river, known to be an important nursery area for anchovy. First results with quad spectrometer do not allow detection of significant concentration in heavy metals, and reveal that environmental signals is mostly hidden by physiological processes for other elements. However, classification of element signal along life trajectories individuals resulted in distinction of several groups with different space-time distribution, a sound basis for studying sub-populations or different cohorts life histories for these two species in the Bay of Biscay.

Abstract reference: IIID_Swearer_05

Realizing connectivity- the influence of early life history on the dynamics of marine metapopulations

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The replenishment of benthic marine populations is the culmination of many processes that influence the production, dispersal, settlement and survival of larvae to maturity. Although there have been recent advances in our understanding of dispersal and its importance to population

connectivity, to date no study has attempted to evaluate all early life-history processes to assess their relevance to the maintenance of marine metapopulations. Using a model temperate reef fish species, the southern hulafish (*Trachinops caudimaculatus*), we present data on larval vertical distributions, larval settlement behaviour from choice experiments, and spatial population structure and integrate these empirical results into a coupled biophysical connectivity framework. We then compare modelled estimates of connectivity to larval dispersal patterns from otolith microchemistry. Our findings reveal that realistic estimates of connectivity depend on a high degree of biological complexity, highlighting the importance of empirical validation of biophysical models of larval dispersal

Abstract reference: IIID_Padilla_06

Using the strontium isotope composition of otoliths from Bering cisco (*Coregonus laurettae*) to determine commercial stock composition in Alaska, USA

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The use strontium isotope composition ($^{87}\text{Sr}/^{86}\text{Sr}$) of otoliths as markers of stock composition in fisheries is a relatively recent management tool. A commercial fishery targeting the anadromous Bering cisco (*Coregonus laurettae*) is occurring in the Yukon River, Alaska, USA. All three of the known global spawning populations occur within Alaska. We used $^{87}\text{Sr}/^{86}\text{Sr}$ values from the freshwater region of otoliths from Bering cisco, from spawning adults of known origin (n= 82), to create a baseline. A 10-fold cross validated discriminant function analysis (DFA) of the three baseline population $^{87}\text{Sr}/^{86}\text{Sr}$ values (Yukon River, n=27; Kuskokwim River, n =25; and Susitna River, n=30) correctly reclassified 97.6% of the fish analyzed. The baseline DFA model was then used to test the commercial samples (n=137). The majority of the commercial samples, with >90% posterior probability, were Yukon River origin Bering cisco (n=134). However, 0.7% (n=1), and 1.4% (n=2) of the commercial samples were classified as Kuskokwim River and Susitna River origin, respectively. The presence of $^{87}\text{Sr}/^{86}\text{Sr}$ values consistent with the Susitna River discovered in the Yukon River baseline (n=1) and commercial samples (n=2) suggests additional spawning areas within the Yukon River or straying among populations.

Abstract reference: IIID_Batista da Silva_07

Preliminary results on the age and growth of yellowfin tuna (*Thunnus albacares*) from western equatorial Atlantic Ocean, based on otolith daily microincrements

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The present work aims to study the age and growth of yellowfin tuna, *Thunnus albacares*, one of

the target species in the fishery for tunas in associated schools in the western equatorial Atlantic Ocean. For this purpose, 118 otoliths were collected in nine fishing cruises in the period from February, 2011 to December, 2012. In the deck, the fish sex was identified, the fork length (FL) were measured by a caliper and the pair of sagittal otoliths were removed, rinsed and dried, identified and stored in plastic bags to be transported to laboratory. Furthermore, the otoliths were embedded in polyester resin and transverse sections were obtained by a low-speed saw, then were ground with wet sand paper and polished in plates with water and aluminum powder. The age and growth study was performed based on two readings of the otolith daily increments. The paired age and length data were adjusted to von Bertalanffy, Gompertz and Logistic growth models by nonlinear least squares and selected based on their AIC (Akaike 's Information Criterion) values. The likelihood ratio test was performed on the von Bertalanffy growth parameters to investigate sexual dimorphism. The fork length ranged between 45 and 168 cm. The values of L_{∞} were estimated in 166.6 cm (pooled data), 171.14 cm (males) and 155.38 cm (females) and the likelihood ratio test reveals that there is no sexual dimorphism. We can conclude that results provide essential information that will improve the knowledge on the yellowfin tuna growth biology.

Abstract reference: IVD_Calò_08

Competency phase affects patterns of fish early life traits

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Fish larvae at competency stage (*i.e.* when they are ready to settle) can display considerable behavioural and movement capacity potentially influencing their spatial and temporal patterns of settlement and distribution in coastal habitats. With the aim to provide insights about the occurrence and the extent of behaviourally-mediated settlement delay, considered as a proxy of behaviourally mediated processes, we investigated the spatial variability of larval phase duration in the saddled sea bream *Oblada melanura* during the competency phase (considering near-settlement and early post-settlement individuals). To accomplish this task we analysed otolith micro-structure. Days after hatching (DAH) in pre-settlers and pelagic larval duration (PLD) in post-settlers were analysed in 2 zones (each corresponding to ~20 km of coastline) separated by ~100 km, along the coast of the region of Murcia (Spain, West Mediterranean Sea). DAH was, on average, ~2 days shorter than PLD. The overall variability of PLD was found to be statistically higher than DAH variability. No significant variability was detected for DAH at the spatial scale considered, where PLD was found to vary significantly. The outcomes of this study suggest that the competency phase could be affected, more than earlier larval stages, by processes able to 1) influence PLD variability and 2) determine a delay during the selection of suitable settlement habitats. This information could have crucial implications for the development of more accurate models of larval dispersal.

Abstract reference: IVD_Shiao_09

Residence depth of the deep-sea fish revealed by stable isotope file

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The whole-life residence depths of benthic deep-sea fish collected from Taiwan waters were reconstructed by the analysis of otolith microstructure and stable isotopic compositions. Otolith $\delta^{18}\text{O}$ profiles suggested that the fish showed two major migratory life history patterns. Most species that laid pelagic eggs and the viviparous cusk eel *Barathronus maculates* underwent downward migration in a distance more than hundreds or even 1000 m during the early life stage. In contrast, some species laying demersal eggs, e.g. *Alepocephalidae* and *Hoplostethus melanopterus* spent all their life around the depths where they were collected. The timing and distance of downward migration varied among the species. Anguilliform fish showed dramatic sink from the pelagic zones to the deep-sea floors after the metamorphosis from leptocephali to juveniles. Grenadiers, Ophidiidae and other species sank to the sea floors during the larval development to juvenile stage. Adult fish collected from deeper sea floors tended to undergo longer downward migration during the early life stage. Otolith $\delta^{13}\text{C}$ profiles suggested that the fish with a longer migration distance had a higher metabolic rate in their early life-history stages than in the later stages. However, the metabolic rate did not vary for the fish living within a narrow vertical range during their larval to adult stages. In conclusion, ontogenetic vertical migration or residential contingents of deep-sea benthic fishes varied among taxonomy, reproductive style and habitat depths, displaying different adaptations or life strategies to the deep-sea environment.

Abstract reference: IIID_Carbonara_10

Use of otolith macrostructure for the identification of yearly birthday groups of European hake (*Merluccius merluccius* Linnaeus, 1758)

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Tag-recapture experiments on European hake (*Merluccius merluccius* Linnaeus, 1758) have recently highlighted a faster growth rate in the western Mediterranean compared to previous hypotheses based on otolith reading and/or length frequency distribution (LFD) analysis. The study of the otolith macrostructure by marginal analysis of hake sagittae in the Central – Southern Tyrrhenian sea has evidenced that the formation of the translucent zone generally corresponds to winter months. In addition, the frequency distributions of the ring distances from the nucleus show two main peaks for each annual ring, in agreement with the presence of two spawning periods (spring-summer and fall-winter). Thus, at the time of the first transparent ring formation, two groups of individual are recognizable, those hatched in summer (age 0) and those born the previous winter (age 1) matching, in turn, with two different spawning times well documented for European hake. This pattern is recurrent in all the age groups. These identified peaks correspond to the back-

calculated total length of 8.5 and 15.7 cm, respectively for summer and winter hake groups for the first transparent ring, and to 19.7 and 23.2 cm in the case of the second transparent ring. The modes of the LFDs of fish caught in winter are coincident with these back-calculated distances of the transparent rings. Hence, these evidences obtained using otolith macrostructure suggest that the underestimation of the growth could be the consequence of the age overestimation for the “summer hake”. Moreover, the identification of these two birth groups has an important impact in terms of the species ecology and management as a different growth pattern might differently impact the prediction of stock productivity when management measures are applied.

Abstract reference: IIID_Cubillos_11

Outline variability of the sagitta otoliths of the Patagonian grenadier (*Macruronus magellanicus*) along the Chilean Patagonian waters

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The Patagonian grenadier, *Macruronus magellanicus*, has an extensive distribution along the continental shelf and also within interior sea, in which gulfs, channels, and fjords are of particular importance for retention of early life history and juveniles of Patagonian grenadier. With the objective of determining probable variations in the shape of sagitta otoliths of the Patagonian grenadier, seven locations were sampled in the study area. The otolith outline was studied on the basis of elliptic Fourier descriptors. The Component Principal Analysis (CPA), the Linear Discriminant Analysis (LDA), and Multivariate analysis of variance (MANOVA) were applied to detect significant differences among areas. The CPA showed a very well mixed data, with extreme variations given by dorsal, ventral, and posterior part of otoliths. The LDA and MANOVA detected significant differences among zones in the outlines of otoliths. The group coming from the inner estuarine zone of Chiloé and Aysén was significantly different from the rest of the zones representing specimens collected in external waters on the continental shelf. Probably, the Patagonian grenadier is structured in resident and migrants than can be expressed phenotypically through intraspecific variability of otolith shape.

Abstract reference: IIID_Morro_12

Fishery-dependent sampling may bias growth estimates

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Reliable predictions on population dynamics depend, among others, on accurate growth estimations, and the conventional approach for estimating growth consists in fitting the von Bertalanffy growth model to length at the age of capture data (*i.e.*, a single observation per fish).

This method derives a single growth curve for all the population but ignores the ontogenetic path of each fish, and hence the existence of possible individual variability. Therefore, it is possible that this composite curve does not correspond to any particular fish's growth. Specifically, in the case of size-selective fishing gears, the pattern predicted is that fish with larger growth rate will reach earlier the selection threshold of the gear and fish with smaller adult size will survive longer. Therefore, conventional analyses of fisheries-dependent samples may bias growth estimates. Conversely, the size of a specific fish at different ages can be back-calculated from the growth marks laid at the otolith, which may allow the derivation of a growth curve for each fish, and explicitly test the hypothesis of the existence of a relationship between growth rate and age. To test this hypothesis three hundred black scorpion fish (*Scorpaena porcus*) obtained from commercial fishermen coming from a stock that presents no sign of overexploitation. The analysis of the back-calculated size-at-age from the otolith growth marks suggests that the growth rate varies between age classes: older individuals systematically presented lower growth rates. A size-selective process where fish with lower growth rates live longer would explain this pattern. In the case of genetically determined growth rate, the existence of such a differential selective fishing pressure between fish of different size may lead to an irreversible, fisheries-induced evolution that might cause undesirable changes in age- and size-structure, and in the dynamics of the stock.

Abstract reference: IIID_Rolls_13

Life history Ba:Ca profiles reveal plasticity in the early life of the diadromous fish (*Centropomus undecimalis*)

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For many fish species, individuals within a population exhibit differing behaviors with respect to important life history events, potentially resulting in behavioral contingents that have distinct spawning and movement habits. Because conditions during early life may propagate into divergent behaviors in subsequent life stages, methods providing information on the unique experiences of larvae and juveniles could be useful in explaining these divergences. To assess the potential for otolith elemental profiles to provide such information for the Common Snook (*Centropomus undecimalis*), a species with unpredictable spawning and movement characteristics, LA-ICP-MS was used to generate continuous life history Ba:Ca profiles for 56 individuals collected throughout the Tampa Bay estuary, Florida (USA). These profiles revealed significant variability in the types of juvenile habitats settled, as well as in the timing of ontogenetic movements from these habitats. Of the profiles examined, 55% exhibited otolith core signatures characterized by decreased Ba:Ca followed by an increase, providing an indicator of the movement of larvae from high salinity, pre-settlement environments into mesohaline, tidally-influenced juvenile habitats. In contrast, nearly half the Ba:Ca profiles indicated settlement in higher salinity environments, suggesting a high degree of habitat plasticity for juveniles of this species. For fish that settled into mesohaline habitats, decreases in Ba:Ca over the first several years of life signaled the ontogenetic transition out of the juvenile habitat, with the timing of emergence ranging from age-1 to age-3. Information on the experiences of early life and juvenile stages may help to inform whether the occupation of different juvenile habitat types, or the precocious or delayed emergence from those habitats, explain the peculiar spawning and movement habits that occur in this species.

Abstract reference: IIID_Mochioka_15

Estimation of hatching dates of Japanese eel preleptocephali collected in the spawning area

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Sampling surveys for eggs, preleptocephali, leptocephali, and spawning adult eels have revealed that the Japanese eel spawns along the southern part of the West Mariana Ridge during new moon periods during late spring and summer. However, little is known about when the peak of spawning and hatching occur within each new moon period. In this study, we examined the relationships between the hatching date of preleptocephali and the moon period. During the R/V Hakuho Maru cruises in June 2011, and May and June 2012, over 250 specimens of preleptocephali that were morphologically or genetically identified as being the Japanese eel were collected by the 3-m Big-Fish plankton net with 0.5 mm and 0.33 mm mesh. Out of the collected preleptocephali, 149 specimens from 26 sampling stations were selected for studying their otoliths. Saggital otoliths were mounted on a glass slides in Euparal and examined at 1000 x with an optical microscope. The number of rings around the hatching check was counted, and hatching dates were estimated from the catch dates. The water temperature at the likely depth of eel spawning appears to be 23-25°C, and the estimated time after fertilization to hatching based on this is approximately 28 hours. The ages of larvae were from 1+ to 4+ days. This information indicates that over 77% of the larvae had hatched before one to two days before new moon. Therefore, it was determined that hatching out of Japanese eel preleptocephali may occur mainly during the periods with dark nights during new moon periods.

Abstract reference: IIID_Vane_16

Connectivity and habitat preference of the Brazilian weakfish (*Cynoscion acoupa*) by analysis of $\delta^{13}C$ of essential amino acids in otoliths

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The connectivity and habitat preference of the Brazilian weakfish (*Cynoscion acoupa*) are poorly known. Described as the most exploited sciaenid fish along the coast of Brazil by some (Farias et al. 2006; Rodrigues et al. 2008), *C. acoupa* is listed as least of concern on the IUCN Red list. This listing is based on the notion that it uses a variety of estuarine habitats as nursery grounds and that therefore mangrove degradation in the region is not thought to be significantly impacting this species (cited from Chao et al. 2010). However, locating a fish species in different habitats does not imply that it utilizes all these habitats as feeding areas for

juveniles. Our main study, therefore, focuses on studying the habitat preference and connectivity of the *C. acoupa* by means of a new method introduced by McMahon et al. (2011), which use the $\delta^{13}\text{C}$ of essential amino acids (EAAs) in otoliths.

The habitat range of the Brazilian weakfish (*C. acoupa*) occurs along the northeastern coast of Brazil, which has hydrodynamic features that can lead to a distinct chemical composition of the coastal waters. This is due to the nutrient rich Amazon river outflow and associated high primary productivity in the north combined with the South Equatorial Current (SEC) at the southeastern coast. As a preliminary study into the $\delta^{13}\text{C}$ of EAAs in *C. acoupa* otoliths, we analysed juvenile (<1 year) otoliths collected at three locations along the Brazilian coast. This study provides a potential insight into how the different chemical composition of the coastal waters might influence the $\delta^{13}\text{C}$ values of EAAs in otoliths and whether $\delta^{13}\text{C}$ values of EAAs can provide a proxy for fish movement over large distances.

Poster Session (IIIE)

Abstract reference: IIIE_Sheridan_01

A potential method for the age determination of Ireland's Norway lobster (*Nephrops norvegicus*) populations using calcified structures.

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Until recently, it was not thought possible to directly and accurately age crustacean species: permanent growth structures were thought to be lost during the biological process of moulting. However recent research has shown that calcified parts of the eyestalk and gastric mill are retained through moults and are thought to preserve annual growth bands in some species of shrimp, crab and lobster. The ability to directly age crustacea would facilitate biological description of crustacean populations in terms of growth, mortality, reproductive success and other age-structured processes that routinely inform management decisions in exploited fisheries. Sections of the mesocardiac ossicle and the zygocardiac ossicles of the gastric mill from *Nephrops norvegicus* caught around Ireland were found to contain growth increment patterns similar to those seen in fish bones and otoliths. Clear, readable growth increments were resolved through a novel preparation method that involved boiling the gastric mill, drying, resin embedding, sectioning, polishing, acid etching and staining. Whilst incremental growth patterns also occur in the calcified regions of the eyestalk in *Nephrops*, these do not appear to follow the growth increment patterns seen within the ossicles of the gastric mill, instead outnumbering them multiple times, possibly related to moulting frequency. Validation is needed to confirm the periodicity of the growth increments within the calcified structures of this commercially important species. Periodic formation of increments within the gastric mill would allow Irish *Nephrops* populations to be directly aged, with precision and potential biases in age estimation analysed through intra- and inter-reader/reading comparisons of age estimates.

Abstract reference: IIIE_Condini_02

Age and growth of Dusky Grouper (*Epinephelus marginatus*) (Perciformes: Epinephelidae) from its southernmost population in Southwestern Atlantic, with a size comparison between offshore and littoral habitats

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This study evaluates the age and growth of the Dusky Grouper (*Epinephelus marginatus*) population at the offshore Carpinteiro Bank (32°16'S; 52°47'W) in the southwestern Atlantic through otolith analysis. We also evaluated the hypothesis that this offshore habitat represents a superior site for fish growth compared to inshore habitats. Samples consisted of 211 groupers captured by small-

scale fisheries between 2008 and 2011 with total lengths ranging from 150 to 1,160 mm. Otolith growth increments were deposited once per year, and opaque bands formed mostly during the summer, as determined through marginal increment analysis. Ages ranged from 1 to 40 years, with most fish aged between 2–8 years and a mean of 7.4 years (SD = 6.9). Von Bertalanffy growth parameters for pooled sexes were L_{∞} = 900.9 mm, K = 0.129 and t_0 = -1.45. Fish from the offshore habitat were generally older and their mean sizes were significantly larger at ages 3, 4, 5 and 6 than fish from the inshore habitat. Our findings highlight the importance of Carpinteiro Bank as an important growing habitat for the Dusky Grouper at the Southwestern Atlantic and suggest that fishing management measures as catch monitoring and fishing quotas should be implemented to ensure the maintenance of this endangered species in this offshore bank in the next future.

Abstract reference: IIIIE_Rezende_03

Age and Growth of Snapper Species (Lutjanidae) from the Tropical Southwestern Atlantic

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On the Brazilian coast (02°N to 25°S) thirteen snapper species can be found around reef habitats over the continental shelf and on offshore islands and oceanic banks. We here report results of otolith based studies for nine species of commercially exploited snappers, captured at latitudes from 02°S to 12°S. Age determination was possible as for all species a consistent and sequential pattern of opaque and translucent marks was observed on both whole and sectioned otoliths under reflected light. The first opaque mark was easily observed in whole otoliths of eight species but not often in sections, a trend reversed on otolith edges of older individuals, where for all species accurate counts were only possible on sectioned otoliths. For yellowtail, lane and dog snapper MIA reveal that new growth marks are formed in April, May and September, respectively. Matched readings of whole and sectioned otoliths were considered the best data set to use on growth estimations, and were used on dog, yellowtail and mutton snappers. The growth coefficient (K) indicate relative slow growth ($0.14 < K < 0.365$) in relation to the maximum ages, species reach 50% of the L_{inf} before four years old. Gear selectivity and fishing fleet dynamics were the main factors affecting growth estimations as it excludes younger age classes. Two alternatives can be applied to solve this effect, fixing t_0 values to zero or backcalculation of size-at-age, although in this case care should be taken over the precision on growth marks measurements, as opaque bands tend to be wider as diffuse. Yellowtail and Southern-Red-snapper presented maximum ages bellow 20 years and maximum assigned age was 33 years for a mutton snapper. This is the first record of otolith based age studies for many southern Atlantic species, and in spite of low latitudes contrast between bands was enough to allow readings with a IAPE lower than 10%.

Abstract reference: IIIE_Egan_04

Can otolith shape discriminate between regional populations of a widely dispersing New Zealand diadromous fish?

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Otolith shape is an evolving and complementary tool to traditional stock discrimination techniques used by fisheries scientists. The shape of an otolith is a species-specific characteristic that integrates genetic and environmental components which together can be used to identify populations that are spatially or temporally discrete during some stage of their life. *Galaxias maculatus* is a diadromous species that spends between three and six months of its life at sea as developing larvae. The translucent juveniles, (whitebait), returning to settle in coastal rivers are the target of an important multi-stakeholder fishery in New Zealand. Although the fishery is inherently data deficient anecdotal evidence suggests it is in decline. Surprisingly little is known about the early life history of this species despite it being fished for over a century. Although mtDNA studies reveal panmixia, biological differences between populations on timescales relevant to fisheries managers must be identified and incorporated into management and conservation strategies. In addition, the degree of mixing is unclear given the dynamic oceanographic processes around New Zealand and evidence from otolith microchemistry studies suggests that at least some populations may be retained regionally. In this context, this study tests the hypothesis of random dispersal and that regional populations of *G. maculatus* can be differentiated by means of sagittal otolith shape analysis. *G. maculatus* were collected throughout the 2013 fishing season from multiple rivers in four regions of the country that support substantial whitebait fisheries. For each fish, the left sagitta was photographed (sulcus side up) using transmitted light and the right sagitta used for ageing. To avoid size bias, only otoliths from 45-55mm fish (TL) were analysed. A combination of Elliptical Fourier descriptors and shape indices were used to compare fish between regions and preliminary results are illustrated here.

Abstract reference: IIIE_Farias_05

What new insights can otolith microchemistry and shape analysis bring to the black scabbardfish, *Aphanopus carbo*, migratory hypothesis?

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With the increase in deep-sea fish landings, stock assessment is relying on a progressively wider variety of scientific tools that provide new insights on species and population ecology and physiology. The black scabbardfish *Aphanopus carbo* is one of the most important commercial

deep-sea fishes in the NE Atlantic, where one stock is admitted to occur in the whole area. The prevailing hypothesis is that at least part of the population migrates northerly from the spawning grounds in Macaronesia to areas off Iceland and the west of the British Isles for feeding. From there, fish continue a clockwise large scale migration, moving along the Portuguese mainland slope and back to the spawning grounds. To evaluate the plausibility of this hypothesis some studies have already focused on life history traits, genetic structure, and otolith science. The present study aims to understand the black scabbardfish migratory dynamics in the NE Atlantic through the combination of otolith trace element composition and contour shape analysis. Otoliths from Iceland, the west of the British Isles, mainland Portugal, and Madeira were collected. Otolith core element composition corroborated that specimens caught off different locations are born in the same place or in similar conditions. In the otolith edge, Mg/Ca, Zn/Ca and Ba/Ca ratios were not significantly different among sampling locations despite the ontogenic differences among locations. These results do not support a close influence of environment nor of physiological processes on otolith chemical composition. The overall discriminating power of otolith contour shape analysis was low, which sustains the existence of a single population.

Abstract reference: IIIIE_Fatnassi_06

Comparing otolith shape for stock discrimination of *Trachinus draco* (Trachinidae) in Tunisian waters

Fatnassi, M.¹; Mahouachi, N-H.¹; Trojette, M.¹; Marsaoui, B.¹; Ben Alaya, H.¹; Chalah, A.²; Quignard, J-P.³; Trabelsi, M.¹

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Otoliths are dense limestones found in the inner ear of bony fish, including: the sagitta, the lapilli and the asteriscus. Otolith shape has been proven to be specific for the species and, also, for populations. The variation in the shape morphology of the otoliths, are reported to be due to environmental characteristics or genetically induced.

In this study, first in Tunisia, the analysis of otolith shape is studied to separate the analyzed stocks and identify morphological differences of the contour's otoliths, using statistical software. These analyzes are applied on the Sagitta, because it is the biggest.

The variability of the otolith shape contour of *Trachinus draco* (Trachnidae), in Tunisian waters, are analyzed to discriminate two lots sampled from two different sites: Raf-Raf (west coast) and Monastir (eastern coast). The both sites are compared by their peers and sex of the fish.

The contour of the otolith has been shown to be a good tool to differentiate stocks of *Trachinus draco* in Tunisian waters. In this part of the stock discrimination, we can confirm that the otolith is a very simple technique, leading to reliable results similar to those that can be found by genetics.

Abstract reference: IIIE_Messaoud_07

Otolithometric comparative study of two Tunisian lagoons population of *Liza aurata* (Lagoon Ghar El Melh and Ichkeul)

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The study of otoliths is interesting in many ways. Including the analysis of their shape which allows the identification and discrimination of stocks. In this context, all otoliths were analyzed to compare the population of *Liza aurata* of lagoon of Ghar el Melh with that of the Ichkeul lake.

The results showed the existence of two different populations with very significant differences (p-value <0.001). Wilks test confirms this result.

The projection of otoliths of these two populations shows that F1 (Absorption = 63.91%) axis divides the population of Ghar el Melh which is the positive side of the Ichkeul which is the negative side.

Abstract reference: IIIE_Khedher_08

Importance of otolith morphology in the discrimination of two Tunisian populations of *Mugil cephalus* (Sea of Tabarka and Ghar El Melh Lagoon)

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During this decade, there was an important development in the domain of halieutic sciences, established on a major party on technological advances in extracting information from fish's Otoliths. That's why; otolithometry is actually used in the most research. This work is based on the morphological study of otolith of *Mugil cephalus*; fish's which were taken from Ghar El Melh Lagoon and Tabarka Sea.

These Otoliths were studied in the order to illustrate the differences and similarities between two populations by multidimensional analysis and discrimination factorial analysis (D. F. A.) after otolith's photo processing by using Photoshop and Shape Software.

The results show that the two populations are separated by axis F1 (absorption= 47, 66).

For both populations, the right and left otoliths of the two sexes (male and female) are separated by the axis F2 (absorption = 10, 64). Statistical analyzes showed, also, a significant differences between

the two groups of fish (p-value <0.05) differences. This shows the uniformity of sea and lagoon populations.

Abstract reference: IIIIE_Marsaoui_09

Use of the contour shape of otoliths in the discrimination of two batches of sea bream (*Sparus aurata*) wild and farmed fish

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Otoliths are the first calcified formations that appear at the embryonic stage, these calcified have long been the subject of research on the biology of fish because they have the property of finely record the stages of growth of individuals during their life.

In this otolithometric study, Otoliths of Royal sea bream, sampled respectively from the sea of Bizerte and an aquaculture site in the Gulf of Hammamet (Ibn Khair in Nabeul), were collected to illustrate the differences or the similarities between these two batches of fish.

Otoliths have been cleaned and photographed and the photos were, then, subjected to treatments of image by software (Photoshop/Shape) and multivariate statistical analyses.

Concerning indices of forms, the graph, representing the two batches of fish Otoliths, reveals the existence of two distinct populations (total absence of symmetry between wild and cultured populations) ; What is confirmed by statistical tests (the P-value < 0,0001, the Mahalanobis-distance test and confusion matrix etc...). These results can be explained by the difference in living conditions (feeding, growth, habitat, etc...) of the two populations.

Abstract reference: IIIIE_Saygili_09

Comparison of otolith morphometric characteristics of *Spicara maena* in the north Aegean sea and sea of Marmara

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In this study, otolith morfometry of *Spicara maena* and population differences were examined between Sea of Marmara and North Aegean Sea. Samples were collected between February 2013 – October 2013 in the North Aegean Sea (Dikili, Altınoluk, Küçükkuuyu) and Sea of Marmara (Karabiga, Bandırma, Tekirdağ) using long lines and game fishing. Totaly 155 *S. maena* specimens were collected in the North Aegean Sea and 168 *S. maena* specimens were collected in the Sea of Marmara. The coordinates of points on the otolith shape has been marked in image analysis program Imagej1.47v. The discriminant analysis is used to separate the morfometric data, to

examine the relationship between the groups and to separate the stocks. All statistical analysis were performed using SPSS19. Differences of otoliths in the Sea of Marmara and the Aegean Sea were analyzed by the help of t-test. It is seen that there were statistical differences in 5 otolith measurements out of 6. It can be observed that the differences in populations were generally caused by the length and the width of otoliths measured. The result of the discriminant analysis, discriminant functions explained 73.5% of the between group variation for morphometric analysis. And the similarity rates of otolith measurements were 69.8% and 77.4% in the Sea of Marmara and Aegean Sea, respectively. According to these results, we detected that *S. maena* populations may differ between Sea of Marmara and Aegean Sea populations.

Abstract reference: IIIE_Diouf_10

Connectivity between populations of the threatened white grouper (*Epinephelus aeneus*) along the West-African coasts inferred by coupling otolith multi-elemental composition and genetic analyses

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The white grouper *Epinephelus aeneus* has been exploited by both small-scale and industrial fisheries along the Mauritanian-Gambian-Senegalese coasts for a long time but its catches permanently decreased since 30 years, probably caused by both over-exploitation and inadequate management measures due to scarce knowledge on its population regulation. Population connectivity along West-African coasts was inferred among seven locations (two in Mauritania, four in Senegal and one in The Gambia) by otolith multi-elemental composition and genetic analyses. Variations in the chemical composition of white grouper otoliths at different spatial scales were used as natural tags when measuring elemental compositions on the otolith edge and on a transect between the otolith core and the edge using laser ablation inductively coupled plasma mass spectrometry (LA-ICPMS). Thirteen elemental ratios were assessed using calcium as reference for Mg, Mn, Cu, Sr, Ba, Pb, Cr, Fe, Co, Zn, Ce, Pr and Nd. Principal component and random forest analyses on multi-element concentrations on otolith edge revealed slight differences between North and South samples but did not allow to accurately separate them. Elemental compositions of transect revealed also slight differences between individuals but no clear different migratory patterns which was in agreement with known seasonal migration along shores in this area. These results were in agreement with the level of gene flow recovered using a set of seven microsatellites loci indicating a unique panmictic population. However, individuals collected off the Senegalese capital, Dakar, presented a significant level of genetic heterogeneity, which questioned the presence of a resident population at this location. Nevertheless, considering the wide distribution range of the unique migratory population, fishery management of the white grouper should be achieved by a common framework between countries.

Abstract reference: IIIE_Kokkin_11

Consequences of within-lake differences in life-history characteristics of pikeperch

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Pikeperch (*Sander lucioperca*) is among the seven commercially most important fish species in Sweden. Around 90% of the total catch is landed in the three largest lakes, in a fishery that has been expanding in recent years. One of the lakes, Mälaren, is heterogenous and consists of different basins with different physical and chemical characteristics. Since 2010, the pikeperch population has been sampled from different basins, for the purpose of stock analysis. Results show that size-at-age and individual growth may differ among basins in a counter-intuitive manner. This is also manifested in selected indicators for stock status. Possible consequences of differences among basins and alternatives for management is discussed.

Abstract reference: IIIE_Arula_12

Discrimination of herring ecotypes using otolith shape and body morphology in the Baltic Sea

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Herring has been historically the most important commercial fish in the Baltic Sea. There are four ecotypes of herring in the Baltic Sea: autumn spawning sea and gulf herring, and spring spawning sea and gulf herring. These four ecotypes have been previously identified and described based on visual inspection of otolith appearance and structure, resulting in discrimination of a number of potential natural populations. Much of this work was done in the 1950s-1970s, and very little progress has occurred since then. The available information level on these four ecotypes is variable. However, it is known that spring herring is nowadays dominating while autumn spawners fall into deep depression in late 1970s and haven't recovered as yet. Some signs of autumn herring recovery can be observed during recent years. Applying formerly used otolith meristics in discrimination of autumn spawning herring of the Gulf of Riga revealed that historical otolith descriptors are not valid anymore. Thus, discrimination of autumn and spring herring by visual otolith appearance is uncertain. Therefore, we have started to apply otolith morphometric image analysis to identify the otolith key to discriminate different herring ecotypes. This information will feed into development of identification guidelines of herring, accompanied by other herring characteristics (e.g. body condition factor, stage of gonad development).

Abstract reference: IIIE_Svirgsden_13

Do Eurasian minnows (*Phoxinus phoxinus*) inhabiting brackish water enter freshwater to reproduce: evidence from a study on otolith microchemistry

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Eurasian minnow (*Phoxinus phoxinus*) is a small slender schooling freshwater fish species. It has little commercial value, but is ecologically important link in food webs as important prey item for many predatory species. Minnow is considered typically as river and lake species, but in Estonia it inhabits also brackish coastal waters of Baltic Sea. Furthermore, minnows inhabiting coastal sea of West Estonian archipelago (it is absent from streams and rivers of those islands themselves) are so far considered as separate ecomorphs, that resides solely in brackish water and do not enter freshwater streams and rivers. In this study we investigated movement patterns of Eurasian minnows using otolith Sr:Ca values to answer the question whether brackish water inhabiting minnows enter freshwater to reproduce. Fish were collected from four different sites (from two bays and two stream mouths) around Saaremaa Island. Migration patterns were inferred from otolith core-to-edge Sr:Ca profiles. The results indicated that at least three distinct migration patterns exist: 1) seawater residency, 2) fast springtime migrations to freshwater or to bays with lower salinities (areas near stream mouths) and 3) prolonged migrations to freshwater (some cases overwintering in streams). Migration patterns listed above were not evenly distributed among sites and some individuals did not migrate to freshwater in every consecutive year. None of the analysed fish (n=124) showed freshwater residency. The findings of this study demonstrate that much more complex migration patterns exists in brackish water inhabiting minnow population than previously thought and at least some individuals do enter freshwater to reproduce.

Abstract reference: IIIE_Condini_14

Dusky groupers from southern Brazil: can littoral fish groups be supplied by neritic populations?

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The dusky grouper *Epinephelus marginatus* is a fish species with broad distribution, usually found in marine rocky bottoms. It is currently listed as “endangered” by the IUCN Red List. The little available information on its population at south Brazil suggests a neritic habitat (Carpinteiro Bank - CB) as the main growth and spawning ground in that region. We investigated whether dusky groupers caught in the rocky-jetties of Rio Grande (a littoral artificial habitat where no spawning occurs) come from the CB. Thus, we sampled 28 individuals at the littoral rocky-jetties and 40 at the neritic CB, and analyzed their otolith sections with LA-ICPMS (core-to-edge profiles) for $^{86}\text{Sr}:$ ^{43}Ca , $^{138}\text{Ba}:$ ^{43}Ca ratios. Otolith cores (age zero) from both sites presented similar elemental compositions for both elements ($P>0.05$). Elemental measurements from ages 1, 2 and 3 were statistically different between sites.

Abstract reference: IIIIE_Martinho_15

Early life stages of plaice *Pleuronectes platessa* in cold-water nurseries

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Plaice *Pleuronectes platessa* (Linnaeus, 1758) is one of the commercially most important flatfish in the European continent, whose captures have reduced considerably in recent decades due to the high pressure of fisheries. Despite the wide distribution of this species, most studies have focused on the North Sea (the central area of occurrence) and very little information is available concerning the populations living at the northern limit of its distribution. Age determination is essential to study the population dynamics of commercially important species such as plaice, and in the case of juveniles it can be inferred through otolith microstructure analysis. Besides age, otolith microstructure also provides information about the growth of individuals and ontogenic stage duration (larval and metamorphosis), proving to be an efficient tool for elucidating early life stage dynamics. The aim of this paper was to investigate the timing of early life events (spawning, larval, metamorphosis and settlement stage duration) and growth of juvenile plaice in two cold-water nursery areas in northern Norway (Valosen estuary, Storfjord) by using otolith microstructure. Settlement was estimated to have occurred between mid-May and early-September (peak from the end May and mid July) in the Valosen, and between the end of May and early July (peak in June) in the Storfjord. In the Storfjord area, larval and metamorphosis stage duration seemed to be shorter, probably due to a reduced window of opportunity for growth and survival at the northernmost extremes. Post-settlement growth, an important part for the nursery function of a nursery ground, was estimated to be 0.70 mm.day⁻¹ (sd=0.40) and 0.68 mm.day⁻¹ (sd=0.48) for the Valosen and Storfjord samples, respectively. The results demonstrate the influence of latitude in the early life stages of plaice, mainly by the regulatory effect of temperature in the growth and survival of larvae and juveniles.

Abstract reference: IIIIE_Dwyer_16

Effect of ageing errors on population dynamics from analytical assessments for two species of flatfish in the Northwest Atlantic

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Ageing errors have been discovered in two flatfish stocks from the Northwest Atlantic in the past decade. Bomb radiocarbon analysis has been used to validate thin sections for yellowtail flounder (*Limanda ferruginea*) in NAFO Divs. 3LNO and Greenland halibut (*Reinhardtius hippoglossoides*) in SA 2 + 3KLMNO and the conclusion was that both species were underaged using whole otoliths, but to varying degrees. We looked at two different assessment models, an ADAPTive framework VPA

for yellowtail flounder and X-tended Survivors Analysis (XSA) for Greenland halibut, to explore how the model output and diagnostics respond to the new ageing. Biological parameters such as growth rates, length and weight at age as well as age at 50% maturity were examined.

A comparison between biomass and F from some of the formulations of ADAPT with the output of ASPIC, the stock-production model currently used to assess yellowtail flounder, shows the same trends, indicating that the VPA might be a tool useful to assess this stock. Biological parameters from the model output indicate a large change in the perception of population dynamics for this stock, when whole otoliths are used to estimate age. The ageing errors for Greenland halibut did not indicate that only the oldest fish are underaged and therefore the changes in model output from the current analytical assessment could be used with modifications to the plus group. In this case, the entire model did not have to be changed, but the plus group was lowered. Some information was lost and there were differences in the population estimates at the beginning of the time series, but estimates for the recent time period were similar. Changes in biological parameters were also apparent for this stock, but to a lesser extent.

Abstract reference: IIIE_Rebaya_16

Importance of sagittal otolith shape in discrimination stocks *Liza ramada* two locations (Sea Cap zebib and Dam Mellegue) Tunisia

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The thin-lipped gray mullet, *Liza ramada* is a pelagic species living in different habitats (estuaries, deltas, lagoons brackish and marine, sea). It can survive in extreme conditions of salinity and withstand sudden changes in water quality. Sagittal otoliths are known to exhibit some phenotypic plasticity inter and / or intra-specific and inter-and / or intra-populationel. This property has often been repeated in the discrimination of fish have evolved in various environmental conditions (in relation to the temperature, depth, substrate, etc.) taking into account possible confounding variables (such intrinsic parameters as sex, size, etc.).

This work, the first in Tunisia, is interested in the study of morphological variability of the species otoliths *Liza ramada* inhabiting two different environments (sea water: sea Cape zebib and Freshwater Dam Mellegue). The results are used to draw the following conclusions: (1) The two populations are separated according to F1 (absorption = 43.82%) axis; marine population occupies the positive part of this axis, while the dam is located in the negative part, test (P-value <0.05) proving that they are different. (2) Right-hand otoliths rights and left-hand otoliths are morphologically similar between males and females of the marine environment test (P-value > 0.05). (3) In contrast, the population of the dam, the significant difference is observed between right-hand otoliths of both sexes. The left otoliths are morphologically similar. (4) For each of these populations, left and right otoliths of males are asymmetric (P-value <0.05). It is the same for females.

Abstract reference: IIIE_Dou_17

Effects of elemental concentration and fish growth on strontium and barium incorporation into otoliths of larval and juvenile flounder *Paralichthys olivaceus*

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This study investigated the effects of ambient elemental concentrations, otolith precipitation and fish growth on strontium (Sr) and barium (Ba) incorporation into otoliths of flounder during larval-juvenile stage under laboratory conditions. Flounder larvae (15 day post hatching, dph) were exposed to a series of Sr (1x, 2x, 3x, 4x of ambient level) and Ba (1x, 2x, 4x, 6x of ambient level) concentrations in an orthogonal two-way design and were continuously reared until 101 dph (86 days). Sr:Ca and Ba:Ca ratios in the otoliths of the experimental fish at the end of the experiment was determined using solution ICP-MS. Results showed that Sr:Ca and Ba:Ca ratios in the otoliths were positively correlated with the levels of the respective elements in the rearing water. Partition coefficients of the ambient elements into otoliths were 0.214-0.220 and 0.155-0.162 for Sr:Ca and Ba:Ca, respectively. Ambient Ba concentration did not significantly influence the uptake of Sr into otoliths. Similarly, ambient Sr concentration showed no significant impacts on the Ba:Ca ratios in otoliths. Both somatic growth rate and otolith precipitation rate of the fish significantly affected otolith incorporation of Sr, but they had no significant effects on Ba incorporation into otoliths. These findings suggest that Sr:Ca and Ba:Ca ratios in otoliths, particularly that formed during larval-juvenile stage, can be used as elemental fingerprints to discriminate among geographic stocks of flounder. However, effects of fish growth and otolith precipitation rate on the otolith elemental fingerprinting should be carefully addressed at stock discrimination.

Abstract reference: IIIE_Jurado-Ruzafa_19

First approach to the growth of *Trachurus picturatus* (Bowdich, 1825) from the Canary Islands (Spain)

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A total of 3532 *Trachurus picturatus* caught by artisanal purse-seiners off the Canary Islands were analyzed between March 2005 and March 2006 (10.4-31.9 cm total length). Biological samplings were performed and sagitta otoliths were extracted and examined. Growth parameters were obtained by direct reading of a total of 1562 whole otoliths ($L_{\infty}=34.9$ cm, $k=0.214$ years⁻¹, $t_0=-2.545$ years). No differences were found between sexes (T_2 Hotelling=0.594; $p>0.05$). Precision (CV=7.5%) and age validation were assessed. Age validation was addressed by means of a qualitative and a quantitative analysis. Marginal increment analysis was carried out for a selection of 453 otoliths. Assigning state of the marginal increment (qualitative analysis) was possible for otoliths of age groups 0, I and II. The monthly evolution of the marginal hyaline increment width was based on the age group 0 (quantitative analysis). We conclude that one annulus is yearly deposited, between January and March (when the spawning season occurs).

Abstract reference: IIIE_Ariza_20

Growth And Age Of The Indo-Pacific Lionfish *Pterois Volitans* (Pisces: Scorpaenidae), In Waters Of Venezuelan Central Coast, South Caribbean

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Lionfish, *Pterois volitans*, is from Indo-Pacific Ocean and it is recognized as the most alarm invasive species in Caribbean coral reefs. It was reported for Venezuela on November 2009. The aim of this research was to describe otoliths as well as estimate daily ages and growth of young fish (< 1 year) of a *P. volitans* population from central coast of Venezuela, South Caribbean. Fifty fish were collected (64-172 mm SL) on June 2013. *P. volitans* otoliths are similar to those of other species of the Scorpaenidae family, big sized (2.35 x 1.19 mm to 4.90 x 2.60 mm), oval-shaped with irregular borders. Relationships between SL (standard length) and otolith diameter and wide were positive, significant and fitted to a linear regression model ($R^2=0.903$, $p<0.05$; $R^2=0.894$, $p<0.05$, respectively). This shows a proportional relationship between otolith size and somatic growth of fish. Youngest individuals were 80 days old, under one day assumption of one increment per day. Daily increments wide changed in size through otolith growth axis. Three stages were observed in the growth trajectory of otolith, the first one with 15 concentric narrow increments around the core ($\sim 2 \mu\text{m}$ increment⁻¹), followed by two groups wider than the first. The results suggest that the first two ring groups belong to larval and post-larval stages, while the last one belongs to the post-settlement stages. Growth rates of fish during these stages are around 0.16, 0.96 and 0.52 mm day⁻¹, respectively. These features of fast development, could explain the settlement of this species in South Caribbean region, specifically at Venezuelan Coast.

Abstract reference: IIIE_Lozys_21

Habitat use and migratory behavior of pikeperch *Sander lucioperca* in Lithuanian and Latvian waters as inferred from otolith Sr:Ca ratios

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The habitat use and migratory behavior of pikeperch *Sander lucioperca* were interpreted from otolith Sr:Ca ratios obtained by Electron Probe Micro-Analyzer. The pikeperch collected from the inland Kaunas city water reservoir consistently showed freshwater signature throughout their life. Those collected from the Baltic Sea and the Curonian Lagoon of Lithuania, and the lower reaches of the Daugava River in Latvia showed habitat shifts between fresh water and brackish water. Although pikeperch can use brackish waters, they predominantly resided in freshwaters. Pikeperch

might also be able to spawn in brackish waters since their larvae can adapt to brackish Baltic Sea waters. The frequent movements/migration between freshwater habitats and the Baltic Sea indicated that pikeperch stocks from a given river/estuary system should be considered as a single management unit despite individual differences in migratory behavior.

Abstract reference: IIIIE_Cerna_22

High growth of the Peruvian anchovy (*Engraulis ringens*) in northern Chile, estimated using daily increment of sagittal otoliths in juvenile and adult fish

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The Peruvian anchovy is a resource of a great economic and social importance, whose stock unit distributed between 16°00'S to 24°00'S is shared between Chile and Peru, with landings reaching 400 thousand tons in 2013. Despite their importance several life history traits remains unrevealed for this species. In the present study daily growth patterns of juvenile and adult of the Peruvian anchovy (*Engraulis ringens*) in northern Chile were determined using micro-increments of sagittal otoliths for the recruitment and fishery season of 2009 and 2010. A characteristic feature was the existence of micro-increments very distinctive, through which a complete sequence of micro-increments were obtained from the primordium to otolith edge for juveniles (12 cm TL). The main finding of this study was the high growth of this species during the juvenile stage where specimens with ages between 56 and 77 days reached in average 8 cm in TL. In adult fish age ranged from 154 to 378 days, which did not matched the 2-3 years old assigned using traditional annulus counts. These findings showed that the Peruvian anchovy grew at an average daily growth rate of 1.2 mm d⁻¹ during the first 3-4 months of life. The Von Bertalanfy seasonal parameters estimated through the entire life history showed a high growth with K = 1.1 and mean length at the first year of 16.3 cm TL. Consequently, *E. ringens* in northern Chile seems to maximize growth to reach the asymptotic length up the end of the first year of life, likely at the expense of drastically reducing its growth thereafter for maximizing reproduction.

Abstract reference: IIIIE_Shivute_23

Horse mackerel *Trachurus capensis* age validation in the northern Benguela

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The Horse mackerel *Trachurus capensis* is an important commercial fish resource of Namibia. It has the highest stock volume and catch off all species caught in Namibian waters, however it has a low market value, hence it is the second highest contributor to the fishing industry after hake (*Merluccius capensis* and *Merluccius paradoxus*). The horse mackerel stock is assessed with an age-structured production model. The model requires the input of catch data, commercial catch per unit effort (CPUE) indices, a survey biomass index and proportions of catch-at-age from surveys and

commercial catches. The objective of the paper is to validate the annual occurrence of otolith zones on horse mackerel. Otoliths were collected from monthly commercial and annual survey samples. The marginal increment (MI) was calculated for each month as the total otolith length minus the length of last complete translucent zone and then calculated as a proportion of the total otolith length. The assumption is that the translucent zones are formed in the months in which the marginal increment is the highest. The cyclical occurrence of translucent and opaque zones was described by Edge analysis where the type of edge present on each otolith was noted. The proportion of each edge type against the date of sampling was computed giving evidence of the time of annuli.

Abstract reference: IIIE_Duncan_24

Investigation of the population and sub-population structure of Albacore tuna in the Northeast Atlantic and Mediterranean

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Albacore tuna is an economically important pelagic resource. Previous studies have reported that the North Atlantic and Mediterranean stocks are genetically distinct. There is also evidence of structure at finer spatial and temporal scales within the North Atlantic and within the Mediterranean. Population subunits within a stock may have different life history characteristics (*i.e.* growth, reproduction) and may respond differently to fishery exploitation. Without a clear understanding of the population structure of Albacore tuna, the efficacy of its stock assessments is reduced and the potential to overfish and deplete the stocks is increased. This study uses otoliths of albacore tuna collected from the western and eastern Atlantic and from different areas within the Mediterranean between 2005 and 2014 to investigate if sub-groups with distinct life histories exist within the stocks. The increment pattern at the larval core of juvenile (ages 2 and 3) and adult (ages 5+) otoliths were interpreted to determine if albacore collected from different areas and at different times during the fishing season have a distinct larval history. Geographic variation in otolith shape, which may reflect differences in life histories and migration pathways, was also analysed using elliptical Fourier descriptors. The results provide insight into the stock structure of albacore tuna which can help to improve the reliability of the stock assessments and the management of the stocks.

Abstract reference: IIIE_Turner_25

Juvenile river herring habitat use and emigration trends throughout their U.S. ranges

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Early life habitat use and movements of successfully recruited adult fish can provide insight into critical nursery habitats. Retrospective analysis of adult otolith chemistry combined with fish-otolith growth models were used to assess nursery habitat selection and emigration size for anadromous alewife (*Alosa pseudoharengus*) and blueback herring (*A. aestivalis*) from 20 rivers in the eastern United States. Freshwater nursery use was observed approximately 8% more frequently in blueback herring than alewives, and use of combined freshwater and estuarine nurseries was observed approximately 9% more frequently in alewife than blueback herring. Estuarine nursery use was more common in southern rivers for both species. Emigration sizes were related to the latitude at the river mouth, watershed area, accessible river kilometers, and the percentage of the watershed in urban use. Principal component analysis was performed on the watershed variables; the first component, which encompassed 46.2% of the variation among rivers, explained 32.0% and 56.9% of the variation in mean emigration size among alewife and blueback herring populations, respectively.

Abstract reference: IIIIE_Yang_26

Life history of three conger eel species in the waters of Northeast Taiwan

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Present study aimed to investigate the life history of conger eel in the northeastern Taiwan by analyzing otolith microstructure and microchemistry. Monthly samples including *Bathycongrus wallacei*, *Gnathophis heterognathos*, and *Uroconger lepturus* were collected from August 2011 to September 2012 from Daxi and Nanfangao fishing harbor. First, otolith microstructure and daily growth increment in the zones of the core, transition zone and accessory growth centre were examined by scanning electron microscope. Durations of metamorphoses were estimated to be, 123 ~ 173 days for *B. wallacei*, 103 ~ 133 days for *G. heterognathos*, and 84 ~ 128 days for *U. lepturus*. The monthly change of the marginal increment ratio indicated that opaque zones on otolith were formed during January and February and the annulus is formed yearly. Yearly age and growth rate were calculated from otolith and data were fitted to von Bertalanffy growth equation. The growth parameters for *B. wallacei* is $L_{\infty} = 886.60$ mm, $k = 0.03$ yr⁻¹, $t_0 = -2.76$ yrs; *G. heterognathos* is $L_{\infty} = 886.60$ mm, $k = 0.03$ yr⁻¹, $t_0 = -2.76$ yrs; and *U. lepturus* is $L_{\infty} = 748.37$ mm, $k = 0.02$ yr⁻¹, $t_0 = -5.82$ yrs. Electron Prob Microanalyzer (EPMA) were used to analyze otolith Sr:Ca ratios of the three conger eels. The result of the Sr:Ca ratio was significantly variated at different life stage of the three eels, with highest ratio at embryonic stage, gradually decreasing at leptocephalus, and remaining constantly low during juvenile and adult stages. Physiology change during early life stage caused the fluctuating in otolith Sr:Ca ratio, and the constantly low Sr:Ca ratios after juvenile stage indicated that the conger eel did not migrate to estuary or fresh water.

Abstract reference: IIIE_Santos-Cruz_28

Morphometric description of sagittae otoliths to differentiate two morphotypes of white mullet *Mugil curema* (Mugilidae) in the Santos bay-estuary complex (Brazil)

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The Santos Bay-Estuary complex, located on the coast of São Paulo State, Brazil, is in a critical state of environmental degradation. The Mugilidae are one of the most exploited families by coastal and estuarine communities along the Brazilian coast. Studies revealed that the white mullet *Mugil curema* is among the major fishery resources of the local artisanal fishery and, the presence of a possible second morphotype was recorded during collection. To distinguish the morphotypes, 304 sagittae otoliths (169 males and 135 females) were used, with ranging between 116 and 423 mm in total length. To test for the hypothesis of two distinct morphotypes, otoliths were described by bivariate morphometrics. 15 morphometric dimensions were measured: length and height of the otolith "LO, HO"; sulcus acousticus length "SAL"; length and height of the cauda "LC, HC"; length and height of the colo "Lcl, Hcl"; length and height of the ostium "Los, Hos"; dorsal and ventral height "DH, VH"; posterior area length "PAL"; length and height of the excisura "LE, HE" and otolith thickness "OT". In multivariate analysis 20 ratios between morphometric measurements were used (LOxHO, LOxSAL, LOxLC, LOxLcl, LOxLos, LOxHC, LOxHcl, LOxHos, LOxDH, LOxVH, LOxPAL, LOxOT, HOxSAL, HOxHC, HOxHcl, HOxHos, HOxDH, HOxPAL, HOxVH and HOxOT) to investigate patterns of otoliths shape. Five groups were formed by cluster analysis by the Ward connection method with 0.667215 cophenetic correlation and Group 3 formed only by individuals of the second morphotype already identified by qualitative morphological description. In the PCA analysis, 7 principal components were identified through the Broken-Stick method, LOxHC, LOxHcl, LOxSAL, HOxOT, HOxHos, LOxDH, LOxPAL and HOxDH, 4 axes demonstrated relationship with the otoliths height, except LOxSAL and LOxPAL, as the larger the otolith height, the shorter the curvature, the larger the sulcus acousticus length and the shorter the posterior area length.

Abstract reference: IIIE_Ferraton_29

Optimising LA-ICPMS rastering protocols and data reduction procedures to produce otolith micro-chemical signatures allowing robust reconstruction of fish past habitats

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Laser Ablation Inductively Coupled Plasma-Mass Spectrometry (LA-ICPMS) allows obtaining high-resolution images of the changes in elemental composition across the otoliths, thereby potentially providing a detailed follow-up of the successive water masses frequented by the fish. Therefore, it is

currently one of the best analytical methods to identify past habitats from otolith signatures. However, to save analysis time, the laser is frequently operated in "raster" mode, all the multi-elemental concentrations measured along scans going from the core to the edge of the otolith being subsequently interpreted as signatures of the successive habitats inhabited. In addition to an obvious problem of autocorrelation between consecutive (overlapping) measurements, this analytical mode raises many issues with regards data pre-treatment, especially when the final signatures are to be compared with those gathered by analysing discrete spots on the edge of the otoliths of fish of known origin. Indeed, not only do the two analytical modes ("raster" vs. "spot") differ in their settings, but data reduction involves many successive steps in both cases. They still constitute a great source of variation (and therefore uncertainty) between laboratories. In this context, we developed an automated yet customizable software package in R for pre-processing the raw data gathered by the two LA-ICPMS operating modes. Equipped with a user-friendly interactive visual interface, this software permits instant visualisation of outputs modification as users modify inputs. This allowed direct investigation of the consequences of different user choices at each possible step of data reduction on the final signatures obtained for both analytical modes. Using data acquired with the two methods along the same otolith transects (for several species and ecosystems), we identified the analysis protocols and data reduction procedures minimizing chemical signatures mismatches between them, clarifying the conditions for attaining reliable fish geolocation from multi-elemental otoliths signatures measured with La-ICPMS rastering.

Abstract reference: IIIIE_Karakulak_30

Otolith Characteristics of Annular Sea Bream (*Diplodus annularis*) in northern Aegean Sea (Turkey)

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In this study, the population parameters and the relationship of otolith and body sizes of annular sea bream (*Diplodus annularis*) were investigated. From March, 2004 to February, 2005, a total of 372 specimens were caught by gill and trammel nets (inner mesh sizes 16, 18, 20, 22, 26, 28, 30 and 32 mm (bar length)) at the depths shallower than 30 m, from the northern Aegean Sea (Gökçeada Island). Total length and weight of the fishes were ranged from 7.7 to 17.7 cm (mean 3.02 ± 0.11 cm), and 5.28-86.30 g (mean 24.91 ± 0.91 g) respectively. The length, width, area, thickness and weight of the sagittal otoliths from 264 samples were measured. Length of all otoliths ranged 2.81–7.36 mm, width 1.87–6.12 mm, area 4.31-22.86 mm², thickness 0.58-1.73 mm, and weight 0.0043–0.0409 g. Right and left otoliths measures were evaluated separately. The relationships between the total length and otolith length (TL-OL), total length and otolith width (TL-OWH), total length and otolith weight (TL-OWT), fish weight and otolith weight of annular sea bream were defined by regression analysis. The total length was directly proportional to weight, with a relationship of $W=0.0078TL^{3.271}$ for females, $W=0.0071TL^{3.305}$ for males, and $W=0.0068TL^{3.315}$ for all samples. Positively allometric growth was observed in all individuals. b value showed a similar trend with other researches carried out in Aegean Sea.

Abstract reference: IIIE_Kitchens_31

Otolith chemical signatures discriminate between nursery areas of yellowfin tuna (*Thunnus albacares*) in the Atlantic Ocean

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In this study, the otolith chemistry of young-of-the-year (YOY) yellowfin tuna (*Thunnus albacares*) was examined to determine whether chemical signatures are distinct across different putative spawning areas in the Atlantic Ocean. Yellowfin tuna is a highly migratory species that is currently managed as a single panmictic stock in the Atlantic Ocean; however, our understanding of the migration patterns and connectivity of Atlantic populations is minimal. Analysis of naturally occurring chemical tracers in otoliths provides a valuable means to reconstruct a fish's environmental history and is thus a promising approach for delineating stock structure of Atlantic yellowfin tuna. YOY yellowfin tuna otoliths were collected from 4 locations in the Atlantic Ocean (Gulf of Mexico, Brazil, Martinique, and Gulf of Guinea) in 2012 and 2013 and trace elemental (Li, Mg, Mn, Co, Cu, Sr, Zn, and Ba) and stable isotopic ($\delta^{13}\text{C}$ and $\delta^{18}\text{O}$) analyses were conducted to investigate regional variation in otolith chemical composition. Preliminary results from the first year of our study showed that otolith chemical signatures varied significantly ($p < .05$) between nursery areas, justifying the use of these natural tracers as regional discriminators for yellowfin tuna. In addition, particularly high spatial separation was observed based on eastern Atlantic (Gulf of Guinea) and western Atlantic (Gulf of Mexico + Brazil + Martinique) nursery areas, indicating the approach has promise for distinguishing migrants displaying trans-ocean movement. This research is ongoing and additional year-classes of YOY yellowfin tuna will be sampled at all locations to determine temporal stability of regional signatures. After we establish our baseline of natal signatures, these data will be used to assign adult yellowfin tuna from the Gulf of Mexico to their nursery of origin. Results of this study will ultimately provide an improved understanding of the stock structure and movement of yellowfin tuna in the Atlantic Ocean.

Abstract reference: IIIE_Zanella_32

Otolith morphology and growth of a short-lived freshwater goby

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The Vrgorac goby *Knipowitschia croatica* (Mrakovčić *et al.* 1994) is a critically endangered freshwater goby species that is endemic to the karstic Matica River of Croatia (Mrakovčić *et al.* 2006). The genus *Knipowitschia*, a member of the sand goby group, is characterised by a short life span which rarely exceeds two years. This study gives the first description of the otolith morphology of the Vrgorac goby. Age structure and growth were calculated separately for males and females,

and pooled as the population, to determine the growth model. The high growth coefficient K obtained in both males and females is owing to the short life span of this species. Age readings from otoliths show that both males and females achieve almost maximum body lengths at sexual maturity in their first year of life.

Abstract reference: IIIE_Maciel_33

Otolith relative growth: a tool to identify sexual dimorphism in the Guri Sea Catfish *Genidens genidens*

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One of the most important catfish in the coastal ecosystems of east coast of South America is *Genidens genidens*. In the Guanabara Bay (22°50'S, 43°10'W, Brazil), a complex estuarine system subjected to a lot of human impacts, it is an important fisheries resource. During the reproductive cycle of *G. genidens*, both sexes present different strategies: females spend energy producing big oocytes and, after spawning, males exhibit oral incubation. We hypothesized that these features affect directly fish growth and otolith development. In order to evaluate this hypotheses, sagittal otoliths were removed and measured (length, Lo, mm; height; Ho, mm; thickness, To, mm; weight, Wo, g). The allometric model ($y=ax^b$) were fitted to verify differences in the relative growth (stanzas) and between sexes. The nonlinear iterative least squares method was applied to fit power models (Lt-Lo, Lt-Ho, Lt-To and Lt-Wo) for each sex, compared through confidence intervals. Residual analyses were used to verify different growth phases. Values of coefficient b showed negative allometric growth for linear measurements ($0.530 < b < 0.776$). Models obtained showed that there are differences between females and males: the otoliths of males grow faster in the four measurements analyzed (b values were significantly different for all models). Three different growth phases were detected: for females, the first one smaller than 28 cm Lt, the second one circa 28 and 35 cm Lt and larger than 35 cm. For males, these phases were also recorded, surprisingly 2-3 cm smaller than in females, possibly related to the larger relative growth rates. Our hypothesis was confirmed and in the next steps of our study we will fit polyphasic growth models for these relationships. Otoliths will be examined for age and growth, clarifying the time and the nature of biological events in the life cycle of *G. genidens*.

Otolith's elemental and isotopic profiles of two populations of twaite shad [*Alosa fallax* (Lacépède, 1803)]: insight into habitat use and discriminate stocks.

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The twaite shad, *Alosa fallax* (Lacépède, 1803), is an anadromous clupeid species that was widely distributed throughout Europe. However, as other European diadromous fishes, *A. fallax* populations are in serious decline due to anthropogenic impacts, including overfishing, barriers to the migration in freshwater and habitat loss. A solid knowledge on the distribution during marine phase, individual migration patterns of both adults and juveniles and homing behaviour is essential to an effective management and conservation of *A. fallax*, but such understanding is incomplete. Several studies conducted over the last decades have demonstrated that otolith microchemistry may be a powerful tool to distinguish movements, life histories and natal origins of fish stocks. Nonetheless, few studies were focused on otolith microchemistry of *A. fallax*, despite they are yielding important results for understanding their biology and ecology. In this study, sagittal otoliths from both adults and juveniles collected in Ulla and Minho rivers (NW Iberian Peninsula) were polished until the primordium in order to determined elemental/Ca ratios (Sr:Ca, Ba:Ca, Mn:Ca, Mg:Ca, Li:Ca, Zn:Ca, Cu:Ca, and Rb:Ca) and ⁸⁷Sr:⁸⁶Sr ratio. For the elemental/Ca ratios continuous profiles were performed from the nucleus to the edge while 60 µm thick half-ring were realized around the otolith primordium for the ⁸⁷Sr:⁸⁶Sr ratio to sample only the first weeks following hatching. Water samples were also obtained in both rivers to verify if otolith microchemistry reflects the ambient chemical composition of the water in which the fish resides. Water samples analyses were conducted using a quadrupole ICP-MS while isotopic analyses were performed on a multi-collector ICP-MS. For otolith analyses both ICP-MS were coupled to a high repetition rate infrared femto-second laser ablation system (Alfamet-Novalase). We discuss the results about two main topics: the discrimination of groups in both freshwater and sea and individual migration patterns of both juveniles and adults.

Abstract reference: IIIE_Araya_35

Partial migration in introduced wild chinook salmon (*Oncorhynchus tshawytscha*) of southern Chile

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Partial migration, the incidence of opposing migration behaviors within the same population, has been a key factor in the invasive ecology of Pacific salmon within South America. Here, we examined such life-cycle variation in of an introduced chinook salmon population in the Aysén watershed, one of the largest fjord systems in NW Patagonia. The chinook salmon is the most successful invasive salmonid species in Patagonia and has recently colonized numerous Patagonian watersheds of the Pacific and Atlantic Oceans. Using analyses of fish scales and otolith strontium:calcium ratios, our results suggest the presence of two distinct ecotypes in the chinook population, an ocean type and a stream type, in a 3:2 ratio. The distribution of back-calculated length at the time of emigration from river to marine habitats showed a mode of 14 cm for the ocean ecotype and 30 cm for the stream ecotype. River residence time for the ocean ecotype ranged from 1 to 10 months, while that of the stream ecotype varied between 14 and 20 months. Returning adults reproduced in riverine habitats between August and March, but reproduction by the stream ecotype was limited to the period between October and February. Our results show that exotic chinook salmon populations established in NW Patagonia present a diversity of life-history strategies, which seems to be as large as the ones exhibited by the species in its native distribution range and in other invaded ecosystems. Studies are needed of the cycles of life among chinook salmon in the Aysén watersheds and other invasive populations of chinook salmon, and their impacts on the invaded ecosystem.

Abstract reference: IIIE_Wells_36

Population structure of albacore (*Thunnus alalunga*) in the eastern Pacific Ocean

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North Pacific albacore (*Thunnus alalunga*) are currently managed as a single stock; however, regional differences in growth rates and movement patterns exist in the eastern Pacific Ocean (EPO). The accurate characterization of population structure and stock mixing is critical to effectively manage highly migratory species such as albacore and a suite of techniques have proven

useful including natural tags in otoliths and muscle tissue. As such, the purpose of this study is to examine chemical signatures in otoliths using stable isotopes of $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ and a suite of trace elements (Ba, Li, Mg, Mn, Sr), in addition to stable isotopes of $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in muscle tissue of albacore collected in two regions of the EPO that have shown limited mixing. Regions include a northern region (offshore Oregon and Washington, $> 40^\circ\text{N}$) and southern region (offshore California, $< 40^\circ\text{N}$). Samples from juvenile and sub-adult (ages 1-4) albacore were collected from each region through recreational and commercial fisheries over a five-year period (2009-2013). Otolith $\delta^{13}\text{C}$ and $\delta^{18}\text{O}$ discrimination between regions exceeded 80% classification success each year using similar age-classes with enriched $\delta^{18}\text{O}$ ($\sim 0.5\%$) in otoliths from albacore in the southern region. Incorporation of trace elements (*i.e.*, Ba, Mg) improved discrimination between regions from 90-100% classification success during each year of the study. Regional differences in oceanography such as high riverine input in the northern region and increased salinity in the southern region match the patterns found in otoliths suggesting the chemical composition of the otolith may be a good proxy for physicochemical conditions of the water mass inhabited. Muscle tissue $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values also suggest differences in overall diets and primary carbon sources between regions with depleted values for each from albacore collected in the northern region relative to similarly sized fish from the south. Ultimately, this information will be coupled with both otolith-derived growth rates and electronic tagging data to provide additional information on the stock structure of albacore in the EPO.

Abstract reference: IIIIE_Paillon_37

Quantifying the role of mangroves for the blackspot snapper (*Lutjanus fulviflamma*) by otolith microchemistry and UVC, in a South-Pacific archipelago (New-Caledonia)

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Mangrove forests are one of the world's most threatened tropical ecosystems and are strongly connected to coral reefs as many reef fish species use mangroves as nursery habitats. The permanence of these species population depends on whether mangroves are obligatory, important or accessory juvenile habitats. In this study, we used otolith microchemistry combined with underwater visual censuses (UVC) and mangrove cartography to estimate the importance of mangroves for the Indo-Pacific coral reef fish *Lutjanus fulviflamma* in the archipelago of New Caledonia. Fish were collected in mangrove and reefs at 11 sites around the island. LA-ICP-MS analyses of otolith edge were used to determine multi-element signatures that discriminate reefs and mangroves. These signatures were then used to reconstruct environmental life history of adults collected on the reefs. We found that all adults displayed a mangrove signature in the juvenile part of their otolith. We also found that individuals presented three different patterns of movement between habitats during their life with the most frequent pattern showing returns to mangrove as adult. UVC and cartography of mangroves confirmed these findings. The analysis of 2942 UVC revealed that the species was absent from isolated islands of New Caledonia archipelago where mangroves were absent. Furthermore, the abundance of *L. fulviflamma* in an area is highly correlated to the area of mangrove within this area. These results highlight the importance of mangroves for the life cycle of the blackspot snapper in New Caledonia archipelago. They indicate at the same time that mangrove is an obligatory juvenile habitat for *L. fulviflamma* and emphasize the potential importance of mangrove in the geographic distribution of the species in New Caledonia.

Abstract reference: IIIIE_Tabouret_38

Reading the environment before reading the otolith: a key to tell *Sicyopterus lagocephalus* stories in La Réunion Island rivers

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Tropical insular systems are colonized by diadromous organisms. Species belonging to Gobioidae, especially the Sicydiinae sub-family, are the largest contributors to the diversity of fish communities and have the highest level of endemism in these systems. Sicydiinae are amphidromous. Once hatching in freshwater, free embryos drift downstream towards the sea where larval development occurs. After a pelagic growth phase, post-larvae recruit back to rivers where they can colonize habitats far upstream. Recent studies suggest downstream movements of individuals living upstream. The evidence of such movements is essential for the management of the insular tropical hydrosystems that suffer from anthropogenic pressures, a loss of the ecological corridor and for the species conservation. However the reconstruction of the habitat use and fish movements, mainly based on the interpretation of the otolith elemental composition, must consider three assumptions: 1) habitats in which the fish can move have significantly different chemical composition, 2) signatures remain sufficiently stable over time and 3) the fingerprint record in the otolith reflects the geochemical signature of the water in which the fish resides. In this study, we investigated during one year the variability of the chemical composition waters of the St Etienne River basin in La Réunion Island (West Indian Ocean) with a monthly sampling throughout 12 sites. The elemental composition (Sr:Ca, Ba:Ca, B:Ca...) allowed a clear differentiation of the sampling sites. Considering these differences, we investigated the elemental composition of otolith of *Sicyopterus lagocephalus* (Teleostei:Gobioidae:Sicydiinae) from 9 sites in the same river basin in order to validate the use of elements as habitat markers. These markers were then used to reconstruct the habitat shifts of *S. lagocephalus* from the estuary to upstream habitats and try to clarify the hypothesis of a behaviour plasticity in this system.

Abstract reference: IIIIE_Libungan_39

shapeR: an R package to study otolith shape variation

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Otolith shape analysis is a powerful tool to detect differences among fish populations sampled at small and large geographic scales. Information on fish population structure is important for the understanding of population dynamics, evolutionary processes and conservation of species. Here, we present shapeR, an open source software package written in the programming language R, specifically designed to collect and analyze otolith shape data. shapeR allows comparison among populations of fish within the same species sampled at small and large geographic scales. shapeR

reads images into R, collects the outlines of the otoliths and performs either a Fourier or Wavelet reconstruction on the otolith outlines. Finally, shapeR provides multiple independent Fourier/Wavelet coefficients as well as other basic shape descriptors such as length, height, perimeter and area of the otolith. Using the shape descriptors it is possible to compare otolith shape variation among populations of fish with both uni- and multivariate statistical analysis. Examples will be shown how to use the package from start to end which includes reading images into R, performing quality checks when collecting otolith outlines, visualizing the mean shape of each population and finally how to compare otolith shape variation among fish populations using Canonical Analysis of Principal Coordinates, Linear Discriminant Analysis and Cluster Analysis.

Abstract reference: IIIE_Nash_40

Spatial linkages in the early life history of north eastern Atlantic herring populations across the north of the British Isles.

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There are two distinct ICES management areas (Area IVaNorth and area IV) for autumn spawning herring across the north of the British Isles. During January and February each year an ICES co-ordinated survey samples the over-wintering herring larvae occurring throughout the North Sea and this index is used as a recruitment index for the stock. It has been known for a long time that larvae spawned to the west of the North Sea most probably are advected into the northern North Sea from the adjacent west of Scotland (IVaN) spawning grounds. However, the extent of larval mixing in the North Sea is not known or where this is occurring has not been quantified. Results from a 'back-tracking' experiment of larvae from their capture site suggests the spatial origins of the larvae. Assuming that the otoliths contain a signature of the natal location, the otolith microchemistry was examined to 'suggest' likely clusters of larvae and their potential source of origin. The results confirm that some larvae in the northern North Sea originate from the west of Scotland. This has implications for the estimate of the recruitment index for herring in the North Sea.

Abstract reference: IIIE_Trojette_41

Study of the morphological variation of the otolith of fish (*Scorpaena Porcus*) between a marine environment (Hammam-Lif) and a middle island (Djerba) in Tunisia

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Otoliths are mineralized concretions in the inner ear of teleost fishes. Each bony fish has 3 pairs of otoliths: Saccular and lagenaires Otoliths are used for hearing and twin otolith helps the fish to maintain balance. At the population level, otolith data allow tackling the recruitment, mortality and can reveal the spatio-temporal structuring of stocks and fish populations. The basis of this study is the detection of the polymorphism between the two populations of *Scorpaena porcus*, in the first hand, and between the otoliths of each population, in the other hand. The database is subject to a discriminant analysis (F.D.A.) to illustrate the differences and/or similarities between the observed groups and optimize the variability existing between them. These analyzes showed the presence of two distinct populations. Thus, Symmetries have been revealed between left and right Otoliths for the population of Hammam-Lif, unlike that of Djerba. These results are confirmed by various statistical tests: Distances Mahalanobis P-value for Fisher distances where the distances between pairs of otoliths is either significant (<0.05), case of the island's population (Djerba Island) or significant name (> 0.05) in cases of marine populations (Hammam Lif).

Abstract reference: IIIE_Frouzova_42

The growth of perch (*Perca fluviatilis*) in newly filled post mining lake

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Detailed growth of perch (*Perca fluviatilis*) was studied in newly filled post mining lake in Czech republic – Lake Most. The perch is common but important predatory fish species. It is also popular subject of angling. For backcalculation of its growth otoliths were used. The results showed amazing increase of growth in first 4 years after populating of fish into the lake. The fish growth increased up to 160 mm per year, mainly in the first year of the life. The reason for it was both populating of perch in practically fishless lake and surplus of the nutrients and food. Similar results were not mentioned from classical reservoirs in Czech Republic so far.

Abstract reference: IIIE_Santana_43

Rosangela Lessa

The importance of the otoliths microstructures in the adjustment of parameters and description of growth in fishes

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Studies of age and growth in fish of a particular species may present dissimilarities according to several factors, such as the use of different hard structures, interpretation of the periodicity of rings and geographical differences. When dealing with otoliths, unique hard structure where it is possible observe daily ages, the use of micro and macrostructures can cause problems in relation to the adjustment of growth rates, as well as in the description of age and growth of the species. Microstructures are commonly used in fish species with short life or in early stages of species with long life cycle. In order to show the importance of the analysis of microstructures in species of long life cycle, were studied and compared the results between different studies of growth for the white mullet, *Mugil curema* and the spotted goatfish, *Pseudupeneus maculatus*. With this two species it was possible to verify that the dissimilarities are related to lack of data on the early life stages of fish, causing: (i) high values of average lengths for each age class; (ii) decrease in the value of K, due to the most rapid growth in the initial phase is not computed; (iii) trend of macrostructures overestimation when used in the early stages of growth; (iv) the absence of a preliminary study of periodicity of the microstructures which can corroborate if the first macrostructures are annual or checks. The absence of the larvae or young-of-year individuals and the number of their microstructures may lead to an underestimation of parameters, making inaccurate estimation of growth.

Abstract reference: IIIE_Khemiri_44

The use of otolith shape to determine stock structure of *Sardina pilchardus* and *Engraulis encrasicolus* in Tunisian coasts

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Sardina pilchardus and *Engraulis encrasicolus* are of great economic importance in Tunisia. However, little is known about their stock structure. In this study, we investigated the stock structure of sardine and anchovy in the open sea and the coastal area of the Gulf of Tunis and the Gulf of Gabes by using otolith shape. Otolith shape was determined using Fourier analysis and compared among specimens sampled from different areas with forward stepwise canonical discriminant analysis (CDA). We found significant differences in otolith shape between open sea and inshore small pelagic groups. Furthermore, otolith shape of anchovy collected in the lagoon of

Bizerte and in the continental shelf: Lake of Ichkeul was analysed using the same methods. The results reveal a clear discreteness of the marine group and the continental one. Such information will have major implications for fisheries management of sardine and anchovy in Tunisia.

Abstract reference: IIIE_Mahe_45

The use of otolith shape to evaluate the stock structure of swordfish (*Xiphias gladius*) in the Indian Ocean

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Swordfish (*Xiphias gladius*) is an oceanic-pelagic species currently fully exploited by several fisheries in the Indian Ocean, with suspicion of overexploitation in the southwest, but without a clear understanding of the real stock structure within this Ocean. Population structure of the Indian stock was studied in the western Indian Ocean using 395 individual samples collected from 2009 to 2014. Sagittal otoliths of the fish have been removed and shape analysis performed on these calcified pieces. Otolith morphometrics data and normalized Elliptical Fourier Descriptors (EFDs) were then extracted automatically by the dedicated image-analysis system TNPC. Preliminary, side effect was tested by Redundancy analysis (RDA) combined to permutation tests on 91 individual samples and showed no significant differences in the outline shape between the right and left otoliths. Consequently, 395 sagittal otoliths were used to identify stocks among several geographical areas (La Reunion, Mozambique channel, Rodrigues, South Africa, South Malagasy, Sri Lanka and Thailand) within the Indian Ocean. To investigate variations of otolith shape according to 4 explanatory variables, Principal Components Analysis (PCA) was applied to EFDs and a RDA with permutation tests was used on the first 7 PC selected by the broken-stick model. These tests demonstrated no significant effects, neither by geographical area ($p=0.203$), sampled year ($p=0.505$), or total length ($p=0.092$). Only, fish sex appeared to be significant ($p=0.026$). Regarding the relationship between the ratio otolith length/otolith width and the total length of fish, size effect was neither significant. Furthermore, Linear Discriminant Analysis (LDA) was performed and overall jackknifed classification success reached 30%. Finally, a clustering analyse has been realised using Ward's hierarchical algorithm, which discriminated 3 different groups, however each group was composed by some individual samples from all geographical areas. In conclusion, all these results did not show a clear geographical separation, and this conclusion corroborate the recent genetic analyse at the Indian Ocean scale while identifying only a single swordfish stock component in the Indian Ocean.

Abstract reference: IIIE_Westgaard_46

The use of otoliths as DNA source in population genetics: population structure of European hake by the use of SNPs

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Otoliths have been used as a DNA source for studying fish population structure, primarily to examine temporal variations. Here we validated their use for detecting spatial structure in the most northerly populations of European hake. Single Nucleotide Polymorphism (SNP) genetic markers in DNA extracted from otoliths were used to describe the population structure of fish sampled off the north-western Norwegian coast (n = 79), a west coast fjord (n = 46), the North Sea (n = 79), the Kattegat (n = 96) and the Bay of Biscay (n = 96). Hake from these areas covered a wide length range (14 – 105 cm), with average area specific lengths varying between 26 and 55 cm. The SNP markers in this study were selected among the 395 SNP loci published from the FishPopTrace project (Milano *et al.* 2011). Based upon the recent findings of Milano *et al.* 2014, we selected a subset of 83 SNP markers to be used in this study. Included are both presumably neutral markers (50 loci) and markers reported as outliers (23 loci) (Milano *et al.* 2014). The objective of this study is to provide both information about the population structure of European hake, *i.e.* whether resident fjord populations can be separated from migrating oceanic fish, and to correlate this with environmental variables such as temperature, salinity etc.

Abstract reference: IIIE_Kemp_47

Trace element mapping of Bight redfish otoliths to assess stock structure in Western Australia

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Bight redfish, *Centroberyx gerrardi*, is endemic to southern Australia, with a distribution that extends along the south and lower west coasts of Western Australia into South Australia (>2000 km), and a depth distribution that extends from coastal waters to depths beyond 300 m. The species is targeted by State commercial and recreational fisheries in coastal waters and is also an important component of the Commonwealth Great Australia Bight Trawl Sector that extends from 3 to 200 nautical miles offshore. Understanding the connectivity of this species across its distributional range is important for developing optimal cross-jurisdictional management strategies. Bight redfish is a particularly slow growing, long-lived species (60-70+ years) and has been reported as having high

vulnerability to exploitation. Resolving the stock structure of Bight redfish is, therefore, particularly important for developing a blueprint for species management.

Trace element maps over otolith transverse sections were produced using laser ablation inductively coupled plasma mass spectrometry (Resonetics M-50 193nm ArF excimer laser coupled to an Agilent 7700s quadrupole ICP-MS). Maps were processed using Lolite (Paul et al., 2012) and detection limits are estimated to be sub-ppm for all elements of interest. Simultaneous measurement of Li, Mg, Mn, Zn, Cu, Sr, Ba and Pb yields elemental information over growth bands from the core to rim of the otolith, correlation of which provides stock structure and movement information for Bight redfish that will inform future stock management plans.

Abstract reference: IIIE_Ruas_48

Updating the age and growth of the rough scad, *Trachurus lathami* Nichols, 1920 in the Southwestern Atlantic

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The rough scad, *Trachurus lathami*, is an important resource for purse seine fisheries in the Southwestern Atlantic Ocean, including the Brazilian coast. Particularly since 1994 its landings decreased a lot, lower to 1,000 tons, after periods of almost 8,000 tons/year. The last estimates available about the age and growth of the species date back to the 1970s. In order to update this information, samples obtained through hydroacoustic surveys along the continental shelf (22°S-29°S) between 2008 and 2010 were analyzed. An otolith subsample (10%) of the 1,312 individuals (total length between 27 and 208 mm) obtained was selected and, following the pre-established methodology for the species, digital images of whole otoliths were submitted to three blind readings. The rings were counted and measured from the nucleus to the beginning of a translucent zone, following the posterior axis. The border, whether it was opaque or translucent, was also recorded. A total of 131 saggitae were analyzed, containing up to seven rings. The second one was not considered as an annual ring and its occurrence has never been reported yet, deserving further investigation. The VBGF parameters were fitted by the nonlinear iterative least square method using all lengths available, resulting in $L_{\infty}=230.69\text{mm}$, $K=0.260\text{year}^{-1}$ and $t_0=-0.693\text{years}$. The age of all individuals of the sample ($n=1,312$) varied between zero and eight years, mainly constituted by fish which were three (42%) and four (27%) years old. Average lengths regarding age (into brackets) were: 38mm (zero), 82mm (one), 116mm (two), 142mm (three), 162mm (four), 178mm (five), 190mm (six), 199mm (seven) and 206mm (eight). The results show that the age structure and growth of the *T. lathami* remains similar to that previously described and this is the most recent data for the rough scad in the Southwestern Atlantic.

Abstract reference: IIIE_Moreira_49

Use of otolith shape analysis for population structure study of the coral reef fish *Stegastes fuscus* (Pomacentridae) from coastal islands of south Brazil

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Damselfish, *Stegastes fuscus*, is a common fish species in the Brazilian waters being considered an ecological key species in reef habitats. One hundred and sixty six adults, ranging from 70 to 120 mm of standard length, were collected in April 2013 by spear fishing in six coastal islands located on Cananéia, Paranaguá, Guaratuba and Babitonga bays, southeast Brazil. A combination of otolith shape indices (circularity, aspect ratio, roundness and solidity) and elliptic Fourier descriptors were analysed by standard multivariate statistical procedures. This study intent to investigate whether otolith shape can be used to differentiate *S. fuscus* populations, providing new knowledge to a rational management of this species.

Abstract reference: IIIE_Rutterford_51

Using otoliths to gauge sub-population responses to warming seas

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Several North Sea fish stocks are characterised by geographically discrete sub-populations, which will each experience uniquely different local environmental conditions. Recent studies indicate that some North Sea species may relocate to track preferred temperatures in warming seas, but emerging research suggests distributions can be constrained by depth or oceanographic features meaning that fish are consequently exposed to warmer conditions. The impacts of warming on sub-populations of targeted species should thus be an important consideration when determining how fisheries are managed into the future. Using analysis of otolith growth patterns and size-at-age data from fisheries surveys, we will examine sub-population specific responses of plaice and cod to warming seas in recent decades, and gauge the level to which stock subdivision modulates predicted impacts of warming climate. Findings from this research will help to identify whether sub-populations have differing capacities to respond to warming; a factor largely overlooked in existing studies of marine environmental change.

Vaster than empires....but more fast: large-scale otolith analyses in population and traceability studies (the FishPopTrace collection)

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Otoliths are robust to most fish processing and preservation methods, with minimal storage requirements. While microchemistry (OTOMC) requires advanced preparation and analytical facilities, shape analysis (OTOMP) is relatively inexpensive using readily available tools. Otoliths are easy to collect, and are incorporated into most monitoring programmes. The existence of otolith archives/collections means that these markers can give a unique and powerful view of population biology which is critical for traceability. The collaborative EU project FishPopTrace (FP7-KBBE-2007-1) was the first large-scale effort to study fish populations and traceability tools within a forensic framework. Genetic and otolith-based baseline traceability signatures were developed from analyses of the spatial and temporal structure of cod (*Gadus morhua*), hake (*Merluccius merluccius*), herring (*Clupea harengus*) and sole (*Solea solea*) populations on a pan-European scale. Otoliths from more than 1300 individual fish were analysed in FishPopTrace; they were photographed to give digital images for OTOMP and then cut to give sections for OTOMC using protocols designed for rapid processing of large sample numbers, taking this beyond the “proof-of-concept” level. The extent of population structure detected through otolith data differed between the species, and OTOMC and OTOMP differed in their ability to discriminate the populations. Cod otolith markers clearly discriminated between Barents Sea, Baltic, and Skagerrak and Kattegat fish. Herring otolith markers clearly identified Baltic, Barents Sea, Western British Isles, and North Sea clusters, as well as distinct groups within the North Sea. Sole otolith data distinguished between Atlantic and Mediterranean sole, and between Bay of Biscay and the North Sea within the Atlantic. Hake otolith markers revealed distinct groups within regions, and classification rates in the Atlantic were higher than in the Mediterranean. Overall, both otolith markers discriminated best between populations within regions, with classification success usually >80%. The combination of OTOMP and OTOMC often improved assignment rates.

Whitefish (*Coregonus lavaretus*) groups in the brackish northern Baltic Sea discriminated by otolith elemental bulk analysis

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Otolith chemistry can be used to discriminate sub-populations of fish and reveal fish migration. In the present study, European whitefish (*Coregonus lavaretus*) was sampled from six different latitudinal sites in the northern Baltic Sea (Gulf of Bothnia). The salinity in this long gulf (>700km) increases from 1 to 7‰ in the north-south direction. Four of the sampling sites were in the sea and two in rivers at the Finnish west coast. The whitefish sampled during spawning from two sites in the sea and the two rivers represent homogeneous stocks. Whitefish from a freshwater lake was collected for comparison. Bulk analysis of the otoliths (n=66) was performed with inductively coupled plasma-optical emission spectrometry to investigate whether the sampled whitefish groups express specific habitat-related otolith concentrations of calcium, strontium, barium, zinc, manganese, and iron. While the calcium concentrations were at the same level in otoliths from different whitefish groups in the northern Baltic Sea, the other elements revealed intriguing characteristics. Otolith strontium concentrations were slightly higher at the southern sampling sites compared to northern ones and thereby showing positive association to capture-habitat salinity. On the other hand otolith barium concentrations were higher at northern sampling sites, thereby associating negatively with capture-habitat salinity. Concentrations of strontium and barium in whitefish otoliths from the lake were very low and high, respectively, compared with whitefish otoliths from the sea. Otolith zinc concentrations increased from north to south. Otolith manganese concentrations did not correlate with capture-habitat latitude, but markedly varied between fish groups. Elevated iron concentrations occurred in fish otoliths from the two northernmost sampling sites. One whitefish group had exceptionally weighty otoliths. Our results indicate that otolith elemental characteristics can be used to distinguish between geographically separated whitefish groups in the northern Baltic Sea and map whitefish to particular spawning areas. The study may aid in revealing the migration habits, including feeding areas and spawning rivers, of endangered (HELCOM) river spawning whitefish.

Slideshow Poster Session (IISS)

Abstract reference: IISS_Vasconcelos Filho_01

Ecomorphology of sagitta from *Plagioscion squamosissimus* (Heckel, 1840) a reservoir on Brazilian northeastern

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Plagioscion squamosissimus is scianid from Amazon Basin. It was introduced in the Brazilian northeastern reservoirs decades ago. The first maturation size is around 19 cm total length (TL). The females grow faster than males. The aim of this study was to describe the *P. squamosissimus* sagitta otolith ecomorphology. Individuals were collected on May and August 2010 at Santa Cruz reservoir, in Brazilian northeast. After specimens identification and biometry, the sagitta otoliths were taken, washed, and photographed. The images analyzed at ImageJ software were measured in millimeters (mm) to estimate area (A), perimeter (P), width (W), height (H) and sphericity (SPH%=W/H*100). The results were showed as mean±SD. The portion occupied by the *sulcus acusticus* was calculated in relation to the total surface (S%). Morphological description was done following Assis (2000). The data were analyzed at R and tested to normality (KS; p>0.05) and homogeneity (KW; p>0.05). The otolith symmetry was evaluated to sex and inner ear side by MANOVA. The effect of the fish TL over the other variables was tested by ANCOVA. In all analysis $\alpha=0.05$. The average size of fifty eight fishes captured was 21.09 ± 4.04 cm (TL). The otoliths showed rhomboid and biconvex shapes, rounded and asymmetric borders. The lack of *excisura ostii*, make the *rostrum* and the *antirrostrum* undifferentiated structures. The *sulcus acusticus* is heterosucoid with anterior margin closed. It is descendent with a suprmedian position. There was no significant difference between the side (MANOVA; p=0.879). The female otoliths were bigger than male (MANOVA; p=0.0000). All the measures were related to TL (p<0.05), except SPH ($67.4 \pm 2.3\%$; ANCOVA; p=0.423). S value ($40.04 \pm 4.05\%$) has a significant relation with TL (p=0.0002), despite of the low r^2 value (0.46). Similar results were observed for other sound producers fishes, that are members of Scianidae family.

Abstract reference: IISS_Pilinlovskij_02

Fish-passes in existing dams of Kražantė and Sausdravas rivers and pass reconstruction in river Vilnia, radio telemetry and conventional tagging studies for Atlantic sturgeon *Acipenser oxyrinchus* Mitchill post release migration

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Our work focuses on measuring animal movements in rivers ecosystems using a variety of tagging methodologies. According to the Lithuanian Catalogue of Water Deposits there are: 733 rivers longer than 10 km, which occupy 325,59 km². Based on the data from Oct. 13, 2011, 94

hydroelectric power plants of different capacity have been built and are operational in Lithuania – from 8 to 1,600,000 kW. Lithuanian rivers have 22 fish paths, which are intended to allow migrating fish to reach their spawning grounds in upper rivers. Three fish passes reconstructed under the project "Fish-passes in existing dams of Kražantė and Sausdravas rivers and pass reconstruction in river Vilnia". In year 2010 Lithuanian reintroduction plan of Atlantic sturgeon was accepted, which includes various measures for long-term reintroduction program: artificial breeding and rearing; herd formation for reproduction purposes and annual stocking. In 2012 and 2013 radio telemetry and conventional tagging studies to assess sturgeon post release migration were performed. Telemetry migration data revealed that more than 90 % percents of released juveniles reaches Curonian lagoon in 15-88 day. Up to date 44 specimens were already caught and reported by anglers and commercial fishermen from Lithuania, Russia, Latvia, Estonia and Finland. One tagged specimen was caught in Åland Islands archipelago more than 800 km from release site.

Abstract reference: IIISS_Zuykova_03

Impact of bias in age reading of the Northeast Arctic cod on stock assessment

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The research is based on data of comparative Northeast Arctic (NEA) cod age-readings carried out in 1992-2012 by the two national institutes: Polar Research Institute of Marine Fisheries and Oceanography (PINRO), Murmansk, Russia, and the Institute of Marine Research (IMR), Bergen, Norway. There were 8,184 pairs of NEA cod otoliths read and compared to assess the difference in age determination between the two laboratories during this period. Differences in age determination were revealed between individual age readers and laboratories as well as between initial and repeated age readings of the same reader. Earlier examination of age reading results of NEA cod conducted by the two laboratories demonstrated biases in age determination of some age groups. Fish age determinations are used in most stock assessment models based on cohort analysis. Errors in age determination might affect stock estimates and subsequent management advice. As the NEA cod stock estimates are currently based on joint data from Russia and Norway, it is rather difficult to assess potential effect of systematic age reading errors in the stock estimates. Therefore, two different stock estimates based either on IMR or on PINRO age determinations were performed to find out possible range in stock size and its status.

Abstract reference: IISS_Silva_04

Morphology of the sagittal otolith of the Whitemouth croaker, *Micropogonias furnieri*, and of the Smooth weakfish, *Cynoscion leiarchus*, in the Southwestern Atlantic, Brazil

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This study aims the description of sagitta otolith of two sciaenids, both important fisheries resources for bottom trawlers in the Southwestern Atlantic, in the Brazilian coast. We analyzed 359 otoliths of *Micropogonias furnieri* from the Bay of Ubatuba (23°24'–23°27'S; 45°00'–45°04'W) and 271 otoliths of *Cynoscion leiarchus* from Sepetiba Bay (22°54'–23°04'S; 43°36'–44°00'W). Sagittal otoliths were measured in terms of length (LO, mm), height (HO, mm), thickness (TO, mm) and weighed (WO, g). The allometric model of Huxley ($y=ax^b$) was fitted by the non-linear iterative least squares method among the total length and the otolith measurements and among the otolith measurements themselves. A t-test was employed to verify whether b was isometric or allometric. *M. furnieri* presents a piriform otolith, with smooth and convex anterior and posterior margins, ventral smooth, sinuous or convex, lobed, wavy or irregular dorsal. The relationship LTxLO was isometric ($b=1,003$; $t=0.1606$; $p=0.4363$) and the others tended to allometry ($p<0.05$), positive ($b=1.048$, LTxTO; $b=3.211$, LTxWO) and negative ($b=0.95489$, LTxHO). The regressions among the otolith measurements showed isometry (LOxTO, LOxWO and HOxWO) and the other models were allometric. *C. leiarchus* presents an oval otolith with anterior, posterior and dorsal smooth and convex, ventral slightly tapered. For the Smooth weakfish, fits involving total length showed negative allometry ($0.5361<b<0.9509$ – LTxLO, LTxHO, LTxTO), except for the otolith weight. Among the otolith measurements, only LOxTO and HOxWO relationships presented positive allometry. The otoliths of the sciaenids analyzed have morphologic differences due to their specific constitution and our results describe the pattern related to the fishes in the environments sampled. Models fitted for both species had similar growth pattern, suggesting that this can be a family's feature. Sagittal otoliths of these species are good predictors of different aspects of the life cycle of these species, including their relative growth.

Abstract reference: IISS_Bal_05

Otolith Biometry - Total Length Relationships in the Population of Hazar Bleak, *Alburnus heckeli* (Battalgil, 1943) Inhabiting Lake Hazar, Elazig, Turkey

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In this study, the relationships between otolith biometry-total fish length of 110 Hazar bleak, *Alburnus heckeli* (Battalgil, 1943) (Cyprinidae) specimens from Lake Hazar were examined. Total

length and weight of specimens ranged between 8.8 and 11.0 cm and between 4.4 and 13.80 g, respectively. The length, breadth and weight of otoliths of each specimen were measured which were respectively 1.48-2.86 mm, 1.02-2.43 mm and 0.0005-0.002 g. There was strong relationship between the otolith length and total length of fish.

Abstract reference: IISS_Ching_06

Spatial and temporal variation in statolith elemental signatures of the *Sepioteuthis lessoniana* around northern Taiwan

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The bigfin reef squid, *Sepioteuthis lessoniana*, is a neritic squid widely distributed in the Indo-Pacific region and of interest to fisheries for its high commercial value. Population structure of *S. lessoniana* has been studied around Japan and Australia, while relevant study on this species is scarce around Taiwan. The objectives for this study are, (1) investigate the spatial and temporal variation of growth and maturation of *S. lessoniana* from the two locations; and (2) investigate the spatial and temporal variation of the trace elements in statolith of *S. lessoniana* from two locations. Monthly squid samples were collected from Ho-ping island (north Taiwan, NT) and Ma-kung (Taiwan Strait, TS) from October 2012 to September 2013, and a total of 949 specimens were collected. Size of the squids from Keelung ranged from 30 to 399 mm in mantle length (ML) and 4 to 2565 g in body weight (BW), and the squids from Penghu ranged from 82 to 302 mm in ML and 59 to 1155 g in BW. A subtotal of 620 squids was aged by reading statolith incremental number. The daily age of squids from Keelung ranged from 55-183 days, and from Penghu ranged from 77-186 days. The result of back-calculated hatching month show they hatched almost all year round, except January and December from Keelung. Concentrations of Mg, K, Ca, Fe, Cu, Zn, Sr, Ba and Pb in statolith were measured using inductivity couple plasma mass spectrometry (ICP-MS). Significant spatial variation were existed by indicated from Fe/Ca, Cu/Ca and Sr/Ca ratios, and temporal variation can also be found by using Sr/Ca and Ba/Ca ratios as indicators. Spatial-temporal difference of statolith elemental composition were found *S. lessoniana* population from two locations, therefore it might indicated different migratory route among seasonal and geographical hatching group of *S. lessoniana*.

Abstract reference: IISS_Walsh_07

The development of fish age determination protocols for commercially important New Zealand species

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In 2011 the New Zealand Ministry of Fisheries (now a part of the Ministry for Primary Industries) held a workshop attended by most of the country's foremost fish-ageing experts. The purpose of the workshop was to develop guidelines for species specific fish age determination protocols, intended for all species routinely aged using MPI funding, with the aim of improving reader accuracy and consistency. This workshop resulted in a document entitled "Guidelines for the development of fish age determination protocols" which is available on the internet.

In 2012 the Ministry for Primary Industries commissioned the development of the first age determination protocols for four important New Zealand inshore species: Snapper (*Pagrus auratus*), trevally (*Pseudocaranx dentex*), tarakihi (*Nemadactylus macropterus*), kahawai (*Arripis trutta*). The ageing protocols, published in 2014, provide a practical guide for ageing each of these species. Each ageing protocol includes:

- A history of the ageing of each species in New Zealand.
- A description of current methods used for preparation of otoliths and interpretation of visible zones.
- Processes used deriving final ages.
- The methods used to estimate and present ageing precision and bias.
- Description and use of reference collections.

Abstract reference: IISS_Farias_08

Using otoliths to identify *Aphanopus carbo* and *Aphanopus intermedius*

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Aphanopus represents one of the most important commercial deep-sea teleosts in the NE Atlantic with about nine thousand tons landed annually and with high commercial value. Two species of the genus *Aphanopus*, the black scabbardfish, *Aphanopus carbo*, and the intermediate scabbardfish, *A. intermedius*, have recently been reported to spatially coexist in some areas of the NE Atlantic. In Madeira, these species are landed as a single commercial category, "black scabbardfish", because it is not possible to distinguish them by external visual examination. Meristic characters and genetics have been used to distinguish the species. In the present study, the suitability of using otoliths to identify the species and quantify the historical frequency of occurrence of the two species was tested. Otolith growth rate, otolith elemental composition, and otolith contour shape were used to compare *A. carbo* and *A. intermedius* caught off Madeira. Otolith growth rate was significantly

different from age 6 onwards and trace element composition was significantly different at age 9. No significant differences on otolith contour shape were found between the two species probably because intraspecific variability was too high. This study has demonstrated that otoliths are an efficient tool to separate *A. carbo* and *A. intermedius*, namely based on trace element composition. This study proved that otolith microchemistry and otolith growth rate could be applied to collections of otoliths to recover historical frequency of occurrence of *A. intermedius* in landings from Madeira. These techniques could also be applied for specimens caught around the Azores and the Canaries, where genetic studies have detected that both species coexist.

Theme IV: Individual Indicators

Plenary session (IVA)

Abstract reference: IVA_Grønkjær_Key

Otoliths as Individual Indicators: Exploring the interaction between genes, physiology and the environment.

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Otoliths are remarkable recorders of fish life-histories and environment, and the flight recorder analogy is not undeserved. Otolith research based on the premise that otoliths are unbiased recorders has significantly advanced ecological and environmental research and is a key pillar of fisheries management. On the individual level the incremental structure of the otolith provides information on age and growth that can be interpreted in relation to life-history traits and environmental stressors. Chemical fingerprints can be used to infer movements and habitat use throughout the life of the fish, and recently also as a means of reconstructing the food webs that support the fish. However, there is one important contrast to the flight recorder. Many of the signals recorded in the otolith are influenced by the state of the individual fish – its physiology and ultimately its genetic makeup. This means that the same environmental signal may be recorded differently in the otoliths of two individual fish. An example is the stress marks laid down in response to unfavorable temperatures. In fish the temperature preferences are closely linked to the genotype and vary even within a population. Consequently, exposure to temperatures that produce a stress mark in a “cold” genotype is not necessarily producing a mark in a “warm” genotype. Another example is the isotopic ratio of otolith inorganic carbon, which is influenced by metabolic carbon and will vary not only according to the isotopic ratio of dissolved inorganic carbon in the fish environment, but also in accordance with the size-specific metabolic rate of the individual. While this is potentially annoying, it also opens new research opportunities into physiology and ecology of individual fish. In this talk I will focus on how studies of otolith structure, inorganic and organic chemistry have been used to gain insights into the ecology, environment and life-history of the individual fish. I will also try to point at some of the exciting research opportunities that arise as we explore the potential of otoliths to reveal the interaction between genes, physiology and the environment.

Abstract reference: IVA_Mohan_01

A multi-proxy approach for estimating estuarine immigration using otolith elements and tissue-specific stable isotopes

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A novel multi-proxy approach was developed to estimate estuarine immigration of juvenile Atlantic croaker, *Micropogonias undulatus*, in subtropical estuaries of the western Gulf of Mexico. Juvenile

fish and water samples were collected along a latitudinal gradient that included positive, neutral, and negative estuary types, to test the hypothesis that juvenile fish immigration timing would vary along a climatic gradient. Lifetime otolith elemental transects of Sr:Ca and Ba:Ca were used to detect fish movement across salinity gradients, while tissue-specific stable nitrogen and carbon isotope ratios revealed time since fish switched from offshore to inshore food webs. A controlled diet-switch experiment determined that liver tissue equilibrated with diet 3X faster than muscle, and that isotope turnover was dependent on growth rate. Nitrogen isotopes in both liver and muscle tissues were highly correlated ($r^2=0.98$) and showed clear geographic separation between bays, suggesting that fish had immigrated inshore at least 3 months prior in all bays. Carbon isotopes in muscle and liver tissues were also correlated ($r^2=0.79$), but overlap in $\delta^{13}\text{C}$ values occurred between all bays, potentially indicating early or late migrants in each bay that will be identified with otolith chemistry. We compare fish immigration timing estimates using otolith chemistry and tissue-specific stable isotopes across each estuary, and present a framework highlighting the advantages, complexities and assumptions of this multi-proxy approach.

Abstract reference: IVA_Campana_02

Image-enhanced burns, bomb radiocarbon and microsatellite DNA improve the accuracy and precision of otolith-based age determinations for redfish (*Sebastes* spp.)

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Past attempts to age Deepwater Redfish (*Sebastes mentella*) and Acadian Redfish (*S. fasciatus*) have been stymied by ad hoc and/or invalid ageing methods, the absence of age validation, and the failure to differentiate among species. Here we report substantial improvements in the tried and true “break and burn” method for ageing *Sebastes* spp. through modern sectioning and image enhancement protocols. Otolith halves sectioned with an Isomet saw allowed routine preparation of completely flat surfaces for charring, while imaging and image enhancements significantly improved the visibility of narrow annuli in older fish. Bomb radiocarbon assays of the otolith core confirmed the accuracy of the resulting age determinations. The use of microsatellite DNA to confirm species identity eliminated past confusion due to species mixtures. Age determinations of 1249 redfish from the eastern coast of Canada demonstrated the presence of significant differences in growth rate and longevity both between the two redfish species and among stocks, with a maximum observed longevity of 70 yr. Even within species and stocks, an individual fish with a fork length of 38 cm could be anywhere between 15 and 50 years of age, highlighting a near cessation of somatic growth after sexual maturation. The presence of annulus splitting in many old *S. fasciatus* rendered some age interpretations virtually impossible, demonstrating that even modern ageing methods cannot provide accurate age determinations in all circumstances.

Abstract reference: IVA_Sturrock_03

Does the elemental composition of marine fish blood and otoliths reflect ambient conditions or physiology? Insights from the lab and the ocean

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Otolith microchemistry is being applied to increasingly complex questions concerning environmental histories and migration pathways of wild fish, yet the mechanisms underpinning element incorporation and the relative importance of environmental, physiological and mineralogical pathways remains largely unresolved. To determine the main controls on otolith elemental composition, mature and immature European plaice (*Pleuronectes platessa* L.) were maintained in near-natural conditions for 7 to 12 months, where physiological (length, weight, condition, plasma protein concentrations, female GSI) and environmental (temperature, salinity) variables were tracked alongside seawater, blood plasma, and/or otolith concentrations of Li, Mg, K, Ca, Mn, Cu, Zn, Se, Rb, Sr, Ba and Pb. Few positive correlations were found between otolith, plasma and seawater elemental concentrations or El/Ca ratios. Otolith Sr/Ca ratios were negatively correlated with ambient concentrations and temperature, and explained by a near 1:1 relationship with plasma Sr/Ca ratios, which were in turn modified by growth, condition and blood protein composition. Large differences in plasma total elemental concentrations were not mirrored in the otoliths, apparently 'buffered' by protein binding and normalization to Ca. However, significant sex-specific differences in otolith Li/Ca, K/Ca and Zn/Ca were observed. The Zn, Sr, Cu and Se concentrations in female blood plasma and/or otoliths were correlated with reproductive investment. Interpretation of these findings alongside data collected from archival-tagged wild plaice with known migrations and temperature histories suggests that in adult marine fish, low environmental variability coupled with physiological noise can complicate the interpretation of otolith microchemistry as a natural tag of location or environmental history, but that elemental markers could indicate key life history events.

Abstract reference: IVA_Elking_04

Maternal contribution process of Trace Elements in Striped Bass Otoliths During Early Ontogeny

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Pathways of possible maternal trace element contribution to developing otoliths in Striped Bass

progeny were examined using soft tissue and otoliths. Otoliths and muscle, liver, kidney, and gonadal (ovaries and testes) tissues were taken from 37 Striped Bass adults from 4 rivers (Roanoke, Neuse, Tar and Cape Fear). Soft tissues were analyzed using ICP-OES and the adult otoliths were analyzed using LA-ICP-MS. Elements deposited within the last year of life in adult otoliths were averaged before subjecting to hierarchical cluster analysis. Soft tissue values were then compared to the clusters using a linear discriminant function analysis. Gonads discriminated between adult otolith clusters > 70% of the time indicating that gonadal tissues should be major contributors of trace elements to progeny otoliths. Striped Bass progeny from known mothers were examined to determine the extent and timing of maternal contribution in progeny sagittal otoliths during early ontogeny (embryo, yolk sac, and non-yolk sac larvae). Embryo sagittal otoliths correctly identified the maternal clusters 100% of the time (n=12), yolk sac larvae 75% of the time (n=16), and non-yolk sac larvae only 44.74% of the time (n=76). Progeny otoliths also identified maternal river (Neuse, Roanoke or Tar); embryos classified the river 75% of the time (n=12), yolk sac larvae 87.5% of the time (n=16), and non-yolk sac larvae 44.19% (n=86) of the time. Results of our study validate the hypothesis of maternal contribution and support the hypothesis of trans-generational markers for anadromous and resident fish.

Abstract reference: IVA_Sirot_05

Using otoliths to link population abundance decline to modifications in individual trophic niche and growth in a tropical fish species (*Bairdiella chrysoura* L., Sciaenidae)

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Otoliths provide the opportunity to better understand the biological and ecological mechanisms responsible for fish demographic changes in disturbed environments by allowing investigation of temporal changes in life history traits. Recent findings suggest that they contain reliable information on individual trophic position available through analysis of the isotopic composition ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) of their organic matrix. These recent methodological developments were used here to study mid-term modifications in both trophic niche and growth for an exploited sciaenid (*Bairdiella chrysoura*) population undergoing an abundance decline in the Terminos lagoon (Mexico) through the last 30 years. Modifications of the species' trophic niche were evaluated by comparing the recent (2011) and past (1980) otolith protein signatures in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ of 62 adults. Changes in growth were investigated by comparing the length-at-age relationships modeled for the two periods using otolith growth mark interpretation, but also through the assessment of body condition and lifetime growth rate. Our results showed that although the abundance of *B. chrysoura* in the lagoon has decreased of 60% since 1980, neither population growth pattern nor diet experienced major modification over the studied time period: growth models did not statistically differ and trophic niches largely overlapped. However, early juvenile growth in the lagoon has decreased since 1980, and average body condition is now higher at the adult stage. Moreover, otolith isotopic signatures also indicated a slight expansion of the population's trophic niche since 1980, concomitant with an increase of $\delta^{13}\text{C}$ and a decrease of $\delta^{15}\text{N}$. The possible links between

these observations and their ecological implications on *B. chrysoira* population are discussed in relation with the environmental changes that occurred in the lagoon since 30 years.

Less-than-daily growth increment formation in fish larvae otoliths: Exploring mechanisms with a bioenergetic modelling approach applied to Atlanto-Iberian sardine (*Sardina pilchardus*)

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Estimating age and growth of fish larvae is often required in connectivity and recruitment studies. When validated, daily growth increment formation in fish larvae otoliths allows such estimations of larval age and growth. However, several experimental studies showed that otolith distinguishable increments are not always formed at a daily scale, which biases age and growth reconstruction. Here, to better understand the factors controlling the formation of a fish larvae otolith, we propose to further develop a bioenergetics-modeling framework previously applied to the seasonal formation of fish otoliths. This model couples both the growth of an otolith and its opacity to the metabolism of the organism. The model relies on well-tested properties of the Dynamic Energy Budget (DEB) theory, which predicts individual growth, development and maintenance as a function of food density and temperature conditions. We apply the model to sagittae of Atlanto-Iberian sardine larvae *Sardina pilchardus*. Data on otolith diameter and growth increment numbers, together with individual known age and total length, from larvae reared in laboratory under four different food conditions (high, intermediate, low and starvation conditions, see Ferreira S. *et al.* abstract) showed that increment deposition was significantly less than one per day. We use data at two different food levels (high and low) to calibrate the otolith module. We first test the ability of the model to reproduce the otolith and fish size for the two other food conditions (intermediate and starvation). We then test the assumption that individuals do not deposit sufficient material of contrasted opacity for an increment to be formed at low food levels. Different opacity functions linked to feeding timings and photoperiod are tested to simulate opacity variation of the deposited otolith material. We discuss the use of this model to reconstruct growth and feeding conditions when increments widths are measured.

Chemical signatures in scales reveal estuarine habitat use and trophic shifts of a highly migratory elopiform

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Fish scales are potential analogues to otoliths that may be sampled for chemical signatures to reconstruct habitat usage of mobile species and trophic dynamics. Studying geochemical tracers in scales is a nonlethal method that allows for sampling from catch-and-release fisheries and threatened or endangered species. We conducted analyses of organic ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) and inorganic (Sr:Ca and Ba:Ca) proxies in scales of Atlantic tarpon *Megalops atlanticus* from the Gulf of Mexico and Caribbean to identify movements across estuarine salinity gradients and ontogenetic trophic shifts. This species was chosen because of its socioeconomic and ecological importance, as well as its large scales, which can reach 6 cm in diameter as adults, allowing subsampling of interior increments for individual scales. Analyses were conducted in two ways: 1) paired subsamples were extracted from individual scales at designated locations to incorporate three life history intervals and homogenized for solution-based ICP-MS and isotope ratio MS and 2) embedded scales were cross sectioned to reveal the surficial calcified layer which was then analyzed with laser ablation ICP-MS to produce continuous profiles of element:Ca ratios. Paired homogenized subsamples showed consistent shifts in Sr:Ca, Ba:Ca and $\delta^{13}\text{C}$ that reflected anticipated ontogenetic movements across estuarine salinity gradients. Strong correlations between Sr:Ca and $\delta^{13}\text{C}$ indicated that $\delta^{13}\text{C}$ primarily reflected salinity rather than trophic position. In contrast, $\delta^{15}\text{N}$ was not correlated with salinity proxies, as expected given trophic enrichments regardless of habitat use. Continuous laser ablation profiles provided high-resolution measurements of Sr:Ca and Ba:Ca values from the core (youngest increments) to the edge (oldest increments). Results indicate that individual behavior is highly variable, with certain fish transiting estuarine gradients into oligohaline waters at different life stages. Our findings demonstrate the utility of scales as non-lethal alternatives to otoliths when investigating fish migrations across estuarine gradients.

Parallel Session IVB (Marking, Laboratory, Microstructure)

Abstract reference: IVB_Starrs_01

Mass-marking fish larvae via maternal transmission of enriched stable isotopes

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Mass-marking fish larvae can facilitate studies exploring connectivity and dispersal patterns of early life history phase fishes. Maternal injections of enriched stable isotopes can provide a means of mass-marking fish larvae during the embryonic phase, because the enriched stable isotopes become incorporated into the larval otoliths (transgenerational marking). However, this is a recent technique that has seen limited development and application. We explored the efficacy of transgenerational marking in the Purple-spotted gudgeon (*Mogurnda adspersa*) and Eastern rainbowfish (*Melanotaenia splendida*), and through a meta-analysis, evaluated the effects of this technique on larval fish morphology and growth across a range of marine and freshwater fishes. Transgenerational marking was highly effective, enabling mass-marking of larvae produced by adult females for up to 6 months post-injection. Multiple unique markers could be produced, with minimal impacts on larvae morphology, survival and growth. Our meta-analysis revealed minimal effects on marked larvae, suggesting that this technique may be applied to explore dispersal and connectivity in aquatic ecosystems.

Abstract reference: IVB_Walther_02

Dietary transmission of isotope spikes to otoliths, fin rays and scales: experimental validation and concentration-dependent mixing models

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Artificial chemical marking of calcified structures, such as otoliths, has emerged as a powerful method to assess stocking success and determine connectivity patterns in fresh and marine systems. Although transgenerational and larval immersion methods have been validated, dietary transmission of enriched stable isotopes to calcified structures would allow minimal handling of animals and reduced expense for flow-through systems. A series of experiments were conducted to validate the feasibility of this method. First, we experimentally manipulated ¹³⁷Ba in diets and successfully marked otoliths in red drum *Sciaenops ocellatus* larvae, fingerlings and sub-adults as well as fin rays and scales of sub-adults. After two weeks, marking success of otoliths was 100% for larvae and fingerlings reared at higher temperatures, indicating the need for sufficient growth to occur for consistently detectable marks. Spiked Ba successfully marked fin rays (86% marked) and scales (100% marked) of sub-adults, providing a cost-effective method for marking structures that

can be sampled non-lethally. Second, we manipulated both ^{138}Ba and ^{86}Sr in diets over four weeks and evaluated uptake of each isotope spike into otoliths. Otoliths were successfully marked with Ba isotopes and stabilized after two weeks of exposure, whereas dietary transmission of the Sr isotope spike was not successful. We used a concentration-dependent mixing model to explore the potential influence of relative differences in water and dietary elemental concentration to explain differential marking success of Ba and Sr in marine systems. This mixing model will be useful for future workers to estimate the feasibility of dietary spiking in various systems with divergent water chemistry. Where feasible, dietary marking of calcified structures may be the method of choice where handling must be minimized or water chemistry cannot be manipulated.

Abstract reference: IVB_Irrgeher_03

Sr isotope pattern deconvolution of LA-MC ICP-MS data to detect individual-specific transgenerational marks in freshwater fish otoliths

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Spiking of fish or water with enriched stable (natural) isotopes has developed into an important tool in fish ecology. In order to apply minimum, non-toxic levels of spike, adequate methods for data reduction needed to be developed in order to identify the spike and properly calculate the original spike composition. This comes of special importance if different isotopic compositions of the spike are applied in order to uniquely identify different entities (e.g. differently marked fish of the same species). In order to meet these prerequisites, a full metrological protocol for data processing during isotope pattern deconvolution (IPD) of an enriched Sr isotope double spike as intrinsic marker in a biological system is described for the first time. The method was developed based on data gained during two transgenerational marking studies of fish, where the transfer of a Sr isotope double spike (^{84}Sr and ^{86}Sr) from female spawners of common carp (*Cyprinus carpio* L.) and brown trout (*Salmo trutta* f.f. L.) to the centre of the otoliths of their offspring was studied by (laser ablation) multi collector inductively coupled plasma mass spectrometry ((LA)-MC ICP-MS). Isotope pattern deconvolution, based on multiple linear regression, serves as simplified alternative data processing strategy to double spike isotope dilution calculations. The outstanding advantage of this mathematical tool lies in the possibility of deconvolving the isotope pattern in a spiked sample without knowing the quantities of enriched isotope tracer being incorporated into the natural sample matrix as well as the degree of impurities and species-interconversion (e.g. from sample preparation). Therefore, all applied spikes could be identified even at concentrations down to $10 \mu\text{g kg}^{-1}$ fish. Biological details of this study are given in the presentation by Zitek *et al.*, Transgenerational marking of freshwater fish otoliths using enriched stable Sr isotopes.

Abstract reference: IVB_Wickström_04

Growth of stocked, marked eels compared to natural recruits in different habitats—a “common garden” approach

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The performance of stocked eels (*Anguilla anguilla* (L.)) has been discussed and questioned particularly since the EU Eel Regulation took effect in 2009. Unbiased comparisons in e.g. growth between stocked and naturally recruited eels have been difficult to achieve as there were few tagging and marking methods allowing sufficiently large number of eels to be recaptured and analysed. The use of small inserted PIT-tags may be useful in eels slightly larger than glass eels, while chemical marking can easily be applied to glass eels and elvers in large numbers. As part of the Swedish Eel Management Plan about 2.5 million quarantined glass eels are stocked annually. Furthermore, since 2009 all stocked eels are marked with an introduced ring of strontium in their otoliths. In this study we have recaptured marked eels from four different environments and compared their growth with naturally recruited eels of the same age from the same sites. Both marine and freshwater populations were studied. Our preliminary results show significantly larger length at age in the stocked eels. However, when corrected for their larger size at stocking a difference in growth rate is no longer present. Finally we discuss possible comparative studies and evaluations to be performed in a near future when more marked eels than today have grown to a size range where sampling is more easily done.

Abstract reference: IVB_Stormer_05

Effects of temperature and ration on the otolith to body size relationship in juvenile Chinook salmon: A test of the direct proportionality assumption.

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The examination of daily growth increments in otoliths is commonly used to estimate the somatic growth rate of juvenile fish. This technique assumes that the otolith and body grow in constant proportion, and has been justified by a positive relationship between otolith growth and fish growth. However, if somatic growth varies with changing environmental conditions, does otolith growth vary similarly, or is there a decoupling between otolith development and fish growth? We examined the relationship between the fork length (FL) and otolith length (OL), width (OW) and area (OA) of juvenile Chinook salmon subjected to simulated summer marine conditions consisting of 3 water temperature (cool, warm, hot), food ration (low, medium, high) experimental treatment combinations. Positive relationships were observed between all otolith metrics and fish size within all temperature treatments, and OA best explained the variability in fish FL indicating that the average of size estimates derived from measuring the radius of both the OL and OW may provide a more accurate approximation of back-calculated fish size than an estimate produced from measuring just one of the metrics alone.

An examination of the residuals of the linear regression between otolith metrics and fish size combined across food rations revealed that the regression-predicted sizes of fish were approximately 4-5 mm smaller and 4-7 mm larger than the observed fish sizes in the cool and hot temperature treatments respectively, and was consistent for all otolith metrics. However, residual analysis showed considerable variability across otolith metrics in the otolith size to fish size relationship after pooling the temperature treatments within the 3 food rations. Our results suggest that increasing ocean temperatures may lead to a decoupling of the relationship between otolith development and fish growth, and should be considered when using otoliths to estimate sizes of juvenile Chinook salmon during early marine life.

Abstract reference: IVB_Williams_06

Assessment of otolith morphometrics as proxies for age in F-based assessments of tropical deepwater fisheries

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Deepwater fisheries in tropical and sub-tropical regions of the Pacific Ocean are typically small-scale artisanal and subsistence fisheries, with strong local economic and cultural value in many Pacific Island countries. Deepwater snappers (Subfamily Etelinae) are the main species targeted by these fisheries which provide an important source of protein to Pacific Island countries that are highly dependent on fish for food security. Most species of deepwater snapper are considered vulnerable to exploitation due to their extended longevity (>40 years) and low mortality rates. However, the sustainability of deepwater snapper fisheries in Pacific Island countries remains uncertain because there are limited resources available to collect the required data for comprehensive stock assessments. Reliable estimates of age composition for exploited deepwater snapper populations are lacking primarily because of the lack of skills and resources required for routine age estimation from sectioned otoliths. Development of alternative low-cost approaches to derive estimates of age for deepwater snapper is required. We evaluate the performance of using otolith morphometrics (weight, length, width and thickness) and fish length to obtain estimates of age for the most important target species in these fisheries: *Etelis carbunculus*, *E. coruscans*, *E. marshi* and *Pristipomoides filamentosus*. We then compare age compositions and fishing mortality rates (F) derived from otolith morphometrics with those derived from counts of annual increments in otoliths for each species. In the absence of sufficient resources to section otoliths for age estimation, we propose that otolith morphometrics are used as a proxy for age in F-based assessments of deepwater snapper fisheries in Pacific Island countries.

Abstract reference: IVB_Mille_07

Does diet influence otolith shape?

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A few recent studies highlighted that food quantity influences fish otolith shape. The main objective of this study is to explore the potential influence of fish diet composition on otolith shape, hypothesizing that food composition (vs food quantity) can impact otolith morphogenesis. Nine economically important fish species, seven roundfishes (European sea bass, red mullet, mackerel, red gurnard, sapphire gurnard, whiting and cod) and two flatfishes (European plaice and common sole), were sampled in the Eastern English Channel during IFREMER Channel Ground Fish Survey (CGFS) in October 2009. Variations in otolith outline shape and in diet were described at an intraspecific level. Otolith shape was reconstructed using elliptical Fourier analysis of the outline and diet was determined by stomach content analysis. For each of the 9 species studied, we partitioned variation in otolith shape according to three potential sources: individual state variables (total length, age, sex and sexual maturity status), environmental factors (temperature, salinity, depth) and diet. The rationale was to disentangle the effects of possible confounding factors on otolith morphogenesis, *i.e.*, individual state affecting morphogenesis through ontogenetic processes and environmental conditions affecting individuals' physiology, from the effect of diet. Variation partitioning allowed us to evaluate the respective influence of individual state, environmental conditions and diet on otolith shape and thus on morphogenesis. From these results we could infer whether food composition had possibly a direct influence on otolith morphogenesis.

Abstract reference: IVB_Bardarson_08

Otolith shape differences related to different migrating behavior in Icelandic cod tagged by Data Storage Tags

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Tagging experiments on Atlantic cod (*Gadus morhua*) in the North Atlantic have revealed varying degrees of migratory behavior. In Iceland, different migration behavior is associated with distinct ecotypes of cod that differ with respect to morphology, age at maturity, genetics, depth during spawning and vertical migrations while feeding. For preservation of stock richness and successful management, it is important to be able to identify stock sub-units. In this study we investigate otolith shape as a phenotypic key to distinguish between ecotypes of cod in Icelandic waters. Cluster analysis was applied to DST data from 203 individuals that had been at liberty long enough

for their behavior to be determined from the DSTs temperature and depth profiles. From this, two main behavior types were identified: shallow (Coastal) vs. migrating and deep (Frontal). These two types could further be separated into two subgroups each, resulting in four behavior groups which are named: Coastal, Intermediate Coastal, Intermediate Frontal, and Frontal. The main behavioral differences between these four groups were the time occupying different depth ranges. Of these fish, 86 otoliths were used for shape analysis. The remaining otoliths were either missing or in such a condition that their shape could not be determined. Otolith shape was described using two-dimensional shape characteristics as well as Fast-Fourier Transforms of their outlines. Generalized linear models (GLMs) were used to assess the discriminating efficacy of the otolith shape. High classification success of the GLMs both for the Coastal and Frontal (>90%), and for intermediate groups (>70%) indicates that otolith shape differs between groups of cod with different strategies of migration. Reconstruction, of the average otolith shape of the groups, indicates visible differences, with Frontal fish having longer more slender otoliths. These results support the growing evidence for sub-structure of the cod population in Iceland.

Abstract reference: IVB_Díaz-Gil_09

Otolith shape fluctuating asymmetry: A misconception of its biological relevance?

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For decades, disruption of the symmetry of body structures has been related to underperformance and, hence, to fitness. In fish, this concept coupled with the claimed evidence for otolith fluctuating asymmetry (FA) under conditions of stress has led to the use of FA as a proxy for individual fitness and population success. Interpreting FA of pairs of hard structures, such as otoliths, that reflect the growth and performance of an individual along its life history is especially appealing, and several studies support the hypothesis linking fitness with the amount of FA. Despite this hypothesis, we report on a number of datasets from a wide range of both, wild populations and experimental designs that show no correlation between FA and any of the usual indicators of fitness or between FA and expected stress gradients. Our results suggest that there may be bias in the published literature toward positive relationships and that results that do not support the proposed hypothesis remain unpublished, despite their importance for a broader understanding of the significance of FA. These results might shed light on a well established concept that may not be applicable in all natural and laboratory based scenarios.

Parallel Session IVC (Chemistry, Shape)

Abstract reference: IVC_Woodcock_01

Natal signatures in the calcified structures of the giant Australian cuttlefish: comparing statoliths and cuttlebones

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The effectiveness of otoliths to recreate environmental histories of fish has led to the expansion of analytical techniques to other calcified structures of fish which allows comparisons between structures, as well as different organisms, such as corals and molluscs. Cuttlefish have two primary calcified structures, statoliths, which are the equivalent to otoliths in fish, and the cuttlebone, an aragonite structure used for buoyancy. Laboratory studies have shown statoliths reflect chemical changes in the surrounding environment, similar to otoliths, but similar studies have not been undertaken on cuttlebones. This study investigated whether statoliths and cuttlebones could be used to trace movements and connectivity of the largest cuttlefish species in the world, the giant Australian cuttlefish *Sepia apama*. We aim to determine the presence of a natal signature from pre-hatch embryos collected from an 8 kilometres stretch of a sub tidal rocky reef in the northern Spencer Gulf, South Australia, where this species is known to aggregate annually to breed, by analysing statolith chemistry using laser ablation inductively coupled plasma-mass spectrometry and then comparing patterns to the chemical signatures of returning adults. Furthermore we aim to quantify if the chemical signatures are analogues between the otolith and cuttlebone structure. Chemical profiles will be used to determine if cuttlefish have dispersed from a common source population (the aggregations site) as well as determine the variability in environments they have moved through throughout their lives. This study will help to identify the percentage of aggregating adults which returned, or have come from different breeding areas thereby allowing for better management and protection of the population.

Abstract reference: IVC_Vitale_02

How long is the lifespan of European hake in the Mediterranean? Validating longevity

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Otoliths of European hake (*Merluccius merluccius*) are very difficult to interpret for age-reading because the growth zone structure in the otolith is complicated by numerous sub-annual growth increments (checks). Furthermore, no clear pattern or periodicity in deposition of translucent and opaque zones is recognizable. These difficulties reflect a high variability in growth and as a result

there is a high uncertainty in longevity for this species in literature. In this preliminary study, two opposing and extreme possible age criteria were accomplished for otolith interpretation of the three females of European hake collected from the archive of the IAMC: (i) the first criteria has tried to discriminate the main increments, *i.e.* younger fish scenario (YFS), (ii) the second criteria considered all increments, *i.e.* older fish scenario (OFS). Bomb radiocarbon was measured in the core of European hake otoliths for which there were two extremes of age interpretation. Of the three fish analyzed, the results indicated that YFS was in agreement with the bomb radiocarbon reference record. This approach was possible because of a recent study that provided a valid dissolved inorganic carbon record from a coral in the Ligurian Sea. The most diagnostic length-at-age for these European hake was aligned with the bomb radiocarbon rise period for the coral reference and was 22 years old. This fish was collected in 1985 at a length of 74.5 cm TL. The oldest fish was likely near 25 years old at a length of 88 cm TL. While two of the age estimates were not optimal in terms of alignment with the bomb radiocarbon reference, they still provided a robust maximum possible age and downgraded reliability of OFS scenario as well as of the fast-growing hypothesis for Mediterranean hake derived by tag-recapture method.

Abstract reference: IVC_Baumann_03

Combining otolith microchemistry and microstructure analyses to infer transpacific migration patterns in juvenile Pacific bluefin tuna (*Thunnus orientalis*)

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Juvenile Pacific bluefin tuna (PBFT) are well-known for migrating across the North-Pacific Ocean from their spawning (Chinese, Japanese Seas) to their feedings grounds (California Current Large Marine Ecosystem, CCLME). However, the duration and timing of migrations need to be better understood for improved management. While recent work (“tracer toolbox”; Madigan *et al.* Ecology 2013) focused on better predicting the departure of PBFT from the Western Pacific, here we explore the suitability of combining longitudinal analyses of otolith microstructure and trace elemental composition in ~ age 1 PBFT for inferring the arrival of individuals in the CCLME. We analyzed 25 individual PBFT that were sampled in fall 2012, and based on their size (63-73 cm SL) they were likely recent immigrants to the CCLME. Concentrations of ²⁰Ca, ⁷Li, ²⁴Mg, ⁵⁵Mn, ⁵⁹Co, ⁶⁵Cu, ⁶⁶Zn, ⁸⁸Sr, and ¹³⁷Ba were determined by LA-ICPMS in transversal otolith sections (9-12 rows, triplicate ablations from core to edge, ø50 µm). The same otolith sections were then further polished to enable microstructure analysis and the subsequent digital overlay of age estimates and trace element:Ca ratios. Age estimates ranged from 320 – 498 days post-hatch, and combined elemental profiles suggested a discernible signature (particularly based on Ba, Mg, Li) near the edge of most otoliths, approximately 30-60 days prior to catch. The pattern was consistent with expectations based on chemical differences between open Pacific and upwelling CCLME waters. A more robust interpretation of the ‘entry signal’ in PBFT otoliths will be possible after pending analyses from an outside control group. Some methodological challenges notwithstanding (e.g., temporal resolution, ageing precision, replicate ablations), we present the approach comprises as a useful addition to the tracer toolbox available to researchers aiming to further understand transpacific migration patterns in *T. orientalis*.

Temperature history estimated with oxygen isotopes in otoliths of the Japanese common conger *Conger myriaster* collected from the continental shelf in the East China Sea.

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The common Japanese conger *Conger myriaster* is a benthic marine species distributed widely in shallow to deep (~800 m) coastal waters of Japan from southern Hokkaido to northern Okinawa, off the Korean Peninsula, and in the Bohai, Yellow, and East China seas. *Conger myriaster* is known to spawn near the Kyushu-Palau Ridge and their larvae have been collected offshore in the western North Pacific. However, inshore fishery captures have rarely included matured individuals. Therefore, little is known about the reproductive ecology and spawning migration of *C. myriaster*. Recently, female specimens were collected from the continental shelf in the East China Sea with higher Gonad Somatic Index (GSI) values than that of coastal specimens, indicating the likely onset of maturation. In this study, the biological characteristics such as water temperature history were estimated with oxygen isotopes in otoliths of maturing *C. myriaster* from the East China Sea are reported and aspects of their spawning migration discussed. Our results showed two types of water temperature histories. Some individuals had narrow changes in their water temperature history (about 15 °C), and in contrast, some had wide changes (10 to 25 °C). Considering the benthic ecology of *C. myriaster*, those individuals must have experienced water temperature higher than 20 °C in shallow coastal areas and not in the outer East China Sea. Consequently, this study suggested some *C. myriaster* females that had begun maturation had migrated from inshore to offshore, before beginning their spawning migration.

Abstract reference: IVC_Piercey_05

Investigating diet and movement of cod (*Gadus morhua*) off Newfoundland using $\delta^{13}\text{C}$ of muscle and otolith amino acids

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Cod from North Atlantic Fisheries Organization (NAFO) zones, Hawke Channel (HC) and Flemish Cap (FC), were collected to determine if the $\delta^{13}\text{C}$ of amino acids in muscle can be replicated in otolith protein. If this is possible important past and present dietary as well as nursery habitat information can be obtained.

The bulk $\delta^{13}\text{C}$ of adult cod muscle from both sites, -19.2 per mil, were similar and indicative of diet mixing between capelin and shrimp. Muscle tissue bulk $\delta^{15}\text{N}$ indicated that HC cod were likely feeding higher in food chain than FC cod. In juveniles and adults $\delta^{13}\text{C}$ of amino acids in muscle did not differ between HC and FC indicating a similar food chain base. The depleted nature of the essential amino acids in muscle suggests a marine phytoplankton base.

Obtaining information on the base of the food chain for different life stages of cod within a population using otolith amino acid signatures would provide valuable information on early life habitats. Initial data comparing $\delta^{13}\text{C}$ of amino acids in muscle to otolith protein are in agreement for essential amino acids and most non-essential amino acids except serine. Therefore, most otolith amino acid dietary information is identical to that recorded in muscle for juveniles. The $\delta^{13}\text{C}$ of otolith amino acids in young fish from both sites were similar, confirming the common marine phytoplankton base. Future work will include populations with different nursery habitats as this may be another tool to differentiate populations.

A trend was observed between $\delta^{13}\text{C}$ of juvenile and adult cod muscle amino acids where most amino acids analyzed were enriched for adults. This may be indicative of a changing diet with growth. Future work will evaluate if a changing diet can be captured in the $\delta^{13}\text{C}$ of otolith amino acids by drilling select otolith annuli to analyze.

Abstract reference: IVC_Valladares_06

Otolith examination in the seahorse *Hippocampus guttulatus*

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New insights into certain life-history parameters of seahorses can help the sustainable management of seahorse wild populations, highly necessary due to their current status as endangered species included in the IUCN Red List Category and Criteria since 1996. In spite of few previous seahorse

ageing studies, age and longevity are currently not assessed for many seahorse species. The purposes of the study were to develop an adequate procedure for extracting otoliths in seahorses, validate daily increments formation and evaluate their feasibility for ageing seahorses. We used specimens of seahorses *Hippocampus guttulatus* provided by 'Proyecto Hippocampus' (CSIC, Spain). Otoliths from newborn and juvenile seahorses of known age (n = 23), and adults of unknown age were analysed (n = 8). Due to the small size of otoliths, dissections were carried out under a microscope fitted with polarized light. A detailed methodology for otolith removal and handling in seahorses is provided. Extracted otoliths were examined under a light microscope to identify otolith types and to determine their size and increments. Otolith types (i.e. sagitta, lapillus or asteriscus) were defined considering their shape, structure and size. The otoliths were photographed and measured under a dissecting microscope and an Olympus BX41 microscope including an Olympus DC50 digital camera connected to a computer with analySIS software. A positive linear relationship ($r^2 = 0.91$) was found for otolith size and seahorse standard length. In juvenile seahorses up to 64 days old, a clear correspondence was established between the age and the number of increments counted in asteriscus otolith type, supporting the formation of daily increments in early life stages of seahorses. A preliminary analysis of otoliths in adult seahorses revealed the presence of defined marks that suggests possible annual or seasonal increments. Further investigations must be undertaken to assess the annual increment pattern in adult seahorses.

Abstract reference: IVC_Nava_07

New open access software designed for the recognition of daily microstructures used in ageing fish.

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Ageing methods for most of the exploited fish species are not validated, and consequently age data series often have important gaps and uncertainty manifesting its consequences on age-based fisheries assessment. In this communication, the research multidisciplinary team of researchers from the research fields of fisheries and engineering plans to develop a new tool designed to add efficiency in the weighty task of estimating the age of fishes based on DGI analysis. The technique is based on new computer algorithms that interpret DGI patterns, whether it be in a semi-automatic way or in such a manner that otolith readers are allowed to interact with the system or run automatically when the technique finds determined types of otolith structural patterns considered consistent and reliable. The technique, when registered, will be made available freely to the international scientific community in the open access mode. Although some image analysis software is available in the market nowadays, including some centered in otolith image processing, they all show at least two main restrictions: they do not manage 'live' images which allow to specifically focus determined microstructures at great magnification, and moreover, the high costs of the software. We present here the first results on the development of this forthcoming software which is expected to resolve the before mentioned limitations. As soon as having ready an appropriate and affordable tool to count and measure daily growth increments in an automatic or semi-automatic way, otolith microstructure analysis can have a reliable age estimation method that can be extended and applicable to age adult fish. Although, in its present form the technique is focused in otolith microstructure analysis, further developments of the initiated software development

intends to extend its application to other calcified structures, namely bivalve shells and cephalopod peaks.

Abstract reference: IVC_Schismenou_08

Disentangling the effects of inherent otolith growth and model-simulated ecosystem parameters on the daily growth rate of young anchovies

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European anchovy (*Engraulis encrasicolus*) juveniles were collected from a coastal area in the North Aegean Sea in July 2007, December 2007 and February 2009. Otolith microstructure analysis revealed that anchovies had hatched from April to December. To reconstruct the potential environment that fish experienced during their development we used the output of a coupled 3D hydrodynamic-biogeochemical model (POM-ERSEM) implemented in the North Aegean Sea, which provided daily values of simulated environmental parameters averaged over the broader sampling area for the period January 2007-February 2009. According to the model simulation, fish caught in February grew in unfavourable conditions in terms of temperature, food availability and water stability; however, they were larger at age and more robust, exhibiting wider otolith increments for the period prior to metamorphosis and similar growth rate to fish caught in July. We hypothesized that these fish were fast-growers and survivors of a population that faced a severe size-/growth-selective mortality event. For further understanding of the environmental effects on anchovy daily growth rate we applied generalized additive model (GAM) analysis on the daily increment widths using the daily values of the simulated parameters as explanatory variables. In the model we also incorporated the effect of 'inherent otolith growth' by including the explanatory parameters 'previous increment width' and 'age'. 'Age' along with 'sampling period' comprised an interaction term that accounted for seasonal variations in ontogenetic changes (timing and duration of metamorphosis) and differences in selective mortality among samples. Results indicated that the daily growth rate increased with increasing food availability (mesozooplankton biomass) and revealed an optimal temperature (~24.5 oC) for anchovy growth.

Abstract reference: IVC_Günther_09

Non-linear back-calculation in juvenile Baltic sprat (*Sprattus sprattus*): Insights into a recruitment-critical life stage

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Daily micro-increment patterns on otoliths can be used to reconstruct early growth histories in

marine fishes. Growth histories of successful individuals of a cohort may uncover traits of individuals with a higher survival probability and thus can help to recognize processes acting on recruitment of a fish stock. Such growth histories are often investigated by length back-calculation. In the simplest case, back-calculation assumes a linear relationship between otolith length and fish length. However, the relationship is far more complex and non-linear in many fish species. To keep the bias of a non-linear relationship small, length back-calculations are often restricted to the larval stage. However, recruitment relevant mechanisms can also occur in post-larval life-stages after metamorphosis, where simple back-calculation fails. This is the case in Baltic sprat, a small pelagic clupeoid fish, where processes acting in the post-larval, early juvenile life-stage are supposed to regulate recruitment variability. We developed a non-linear, regression-based length back-calculation algorithm accounting for morphometric changes during ontogeny, to investigate early juvenile growth in sprat. In contrast to linear back-calculation, our method produced higher larval and juvenile growth rates and lower growth rates during metamorphosis. Inter-annual variations in newly back-calculated juvenile growth rates indicate that high growth in this post-larval life-stage correlates with strong year-classes in the Western Baltic Sea. We found that individuals born early in the year display poor growth in the juvenile stage as they experience high water temperatures at a large size requiring high food availability. In contrast, later born individuals experience highest water temperatures at a smaller size with moderate food requirements and lower water temperatures thereafter. In essence, the combination of a cold winter and a warm summer benefit juvenile growth, survival and recruitment by shifting main spawning and post-larval growth later into the summer season.

OtoChuki Session (IVD, Tuesday)

Abstract reference: IVD_Lavergne_08

Early life history of *Terapon jarbua* (Forsskål, 1775) using microstructures and Laser Ablation ICP-MS elemental composition of otoliths

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Early life history of *Terapon jarbua* (Forsskål, 1775) was examined using otolith microstructures, otolith elemental composition and assignment test and was linked with previous analysis of microsatellite genotypes. Eight estuaries from Socotra Island and mainland Yemen were sampled in 2007 and 2008. Laser ablation inductively coupled mass spectrometry transects across otolith growth axes of juvenile *T. jarbua* were used to estimate early life migration of the species. Sr:Ba ratios across each transect were particularly sensitive to transitional environments, such as estuaries. *Terapon jarbua* exhibited a natal marine phase, migrating after a relatively short planktonic larval stage into estuarine systems as post larvae or young juveniles in which they remained for at least two years. An average planktonic larval duration of 25.1 days was estimated and remained stable over the studied region. The study highlighted that elemental fingerprints of otolith edges have great potential for discriminating nursery areas at both small (< 10 km) and large geographical scales (80 % assignment accuracy). This tool might be useful to estimate the contribution of specific estuarine nurseries to any adult stocks and should be used in the future to build pertinent management and conservation programs devoted to the Socotra estuaries. However, temporal variability in otolith elemental composition has been observed, thus making its routine usage more demanding. Elemental fingerprints of the otolith nuclei had meanwhile a lower potential for discriminating unknown marine spawning grounds but pointed out the existence of several different spawning areas, thus supporting previous results on population structure and population dynamics of *T. jarbua*.

OtoChuki Session (IVD, Thursday)

Abstract reference: IVD_Ashworth_01

Are relationships between fish length and otolith radius enhanced by accounting for correlated errors? Implications for back-calculation.

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Sclerochronology in fishes has focused the relationships of the distances between the annually-formed zones in otoliths and certain environmental variables. The results have frequently been used as a proxy for the relationship between somatic growth and those environmental variables. Yet, as somatic and otolith growth may be partially uncoupled, inter-annual variations in the increment widths in otoliths may not fully reflect the trends in somatic growth. Thus, for sclerochronological studies on a species, it is important to determine the precise relationship between otolith and somatic growth. This study has elucidated the extent to which variations in otolith and somatic growth are related in fish of the same age, by deriving the otolith sizes and length (TL) at age at capture for six species whose environments, *i.e.* marine vs estuarine and tropical vs temperate, and life cycle characteristics, *i.e.* gonochoristic vs hermaphroditic, differ and, for one species (*Acanthopagrus butcheri*), different periods and estuaries. A sagittal otolith from each fish was sectioned and the distance from its primordium to the outer edge of the otolith measured to 1 μm along an axis adjacent to the sulcus and perpendicular to the growth zones. The Schnute, rather than the von Bertalanffy growth curve, typically provided a better fit to both the TL and otolith size at age and was thus used for analyses. Growth curves were fitted simultaneously to somatic and otolith data. The deviations from these curves, which were assumed to have a bivariate normal distribution, were positively correlated and differed significantly from zero. Thus, for fish of a given age in a given habitat, the rates of somatic and otolith growth are linearly related. Variations in otolith growth resulting from environmental change - unless markedly outside the range of the data - thus result in a predictable level of change in somatic growth.

Abstract reference: IVD_Morat_02

Variability of the otolith growth rate and age of young-of-the-year chub, *Squalius cephalus* (Linnaeus, 1758), from a hydraulic reach in the Rhône River.

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Understanding the influence of environmental characteristics on the age and growth of young-of-the-year is a fundamental challenge in ecology. Young-of-the-year (YOY) chub (Cyprinidae, *Squalius cephalus*) were collected in the Lower Rhône River (South of France) along the shoreline of a 30-km hydraulic reach. The spatial distribution, age, hatching period and daily growth rate (DGR) of the YOY otolith were investigated to evaluate differences in four contrasted areas along the reach (a by-passed section, tailrace, confluence area and reservoir). The otolith shape analysis clearly distinguished the areas into three groups (the by-passed section, the tailrace and a third group formed by the reservoir and the confluence individuals). The mean age of fish from the medium-size class varied according to areas from 149 days for the tailrace to 166 days for the by-passed section. The estimated hatching period extended from mid-February to the beginning of June with a peak in April and May. The otolith DGR showed similar curve profiles for the four areas with low values during winter and spring ($\sim 1.0 \mu\text{m d}^{-1}$) and a large increase in summer (between $1.9 \mu\text{m d}^{-1}$ and $2.4 \mu\text{m d}^{-1}$ on average). These variations seemed in agreement with the 2013 hydro-climatic context. However, differences in the DGR curve profiles among the areas were observed from August to mid-September. The highest DGR values ($\sim 2.8 \mu\text{m d}^{-1}$) were estimated for YOY of the tailrace and the

lowest for fish in the by-passed section ($\sim 2.1 \mu\text{m d}^{-1}$). These differences could reflect local variations in the habitat conditions.

Abstract reference: IVD_Traczyk_04

Age, growth, and distribution of the Antarctic fish *Pseudochaenichthys georgianus* based on otolith morphometry

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The *Pseudochaenichthys georgianus* were sampled in the summer between 1979 and 1990 at South Georgia Is. The otoliths of *Ps. georgianus* contained microincrements similar to daily increments in the otoliths of fishes from temperate and tropical waters. The estimated age from microincrements was similar to that inferred from otoliths weight and body length distribution. Changes of the microincrements growth pattern are described that appear to be related to larval, hatching and metamorphosis, and have similar pattern to that ones previewed in *Chaenocephalus aceratus* and *Champocephalus gunnarii*. Larvae and postlarvae of that species have similar habitats during early life history, that reflect in their close up otoliths morphology. Average width of daily increments of otolith larvae are: 0.0018 mm for *Ps georgianus*, 0.0015 for *C. gunnarii*, 0.001 for *C. aceratus*, and for their postlarvae as follow: 0.0028, 0.0024, 0.0016 mm. The ages of 300 individuals of *Ps. georgianus* were determined by microincrements. The age of fish sampled by the fishery was predicted from otolith weight frequency with equation: $\text{age}[y]=81.98 \times \text{OW}[g]-0.483$; $R^2=0.97$. For *Ps. georgianus* the von Bertalanffy growth parameters were, $k = 0.28$, $L^\infty = 66.1$, $t_0 = 0.008$, $L_0=0.15$ cm. Otoliths size at age were established.

Abstract reference: IVD_Calò_08

Competency phase affects patterns of fish early life traits

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Fish larvae at competency stage (*i.e.* when they are ready to settle) can display considerable behavioural and movement capacity potentially influencing their spatial and temporal patterns of settlement and distribution in coastal habitats. With the aim to provide insights about the occurrence and the extent of behaviourally-mediated settlement delay, considered as a proxy of behaviourally mediated processes, we investigated the spatial variability of larval phase duration in the saddled sea bream *Oblada melanura* during the competency phase (considering near-settlement and early post-settlement individuals). To accomplish this task we analysed otolith micro-structure. Days after hatching (DAH) in pre-settlers and pelagic larval duration (PLD) in post-settlers were analysed in 2 zones (each corresponding to ~ 20 km of coastline) separated by ~ 100 km, along the coast of the region of Murcia (Spain, West Mediterranean Sea). DAH was, on average,

~2 days shorter than PLD. The overall variability of PLD was found to be statistically higher than DAH variability. No significant variability was detected for DAH at the spatial scale considered, where PLD was found to vary significantly. The outcomes of this study suggest that the competency phase could be affected, more than earlier larval stages, by processes able to 1) influence PLD variability and 2) determine a delay during the selection of suitable settlement habitats. This information could have crucial implications for the development of more accurate models of larval dispersal.

Abstract reference: IVD_Shiao_09

Residence depth of the deep-sea fish revealed by stable isotope file

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The whole-life residence depths of benthic deep-sea fish collected from Taiwan waters were reconstructed by the analysis of otolith microstructure and stable isotopic compositions. Otolith $\delta^{18}\text{O}$ profiles suggested that the fish showed two major migratory life history patterns. Most species that laid pelagic eggs and the viviparous cusk eel *Barathronus maculatus* underwent downward migration in a distance more than hundreds or even 1000 m during the early life stage. In contrast, some species laying demersal eggs, e.g. *Alepocephalidae* and *Hoplostethus melanopterus* spent all their life around the depths where they were collected. The timing and distance of downward migration varied among the species. Anguilliform fish showed dramatic sink from the pelagic zones to the deep-sea floors after the metamorphosis from leptocephali to juveniles. Grenadiers, Ophidiidae and other species sank to the sea floors during the larval development to juvenile stage. Adult fish collected from deeper sea floors tended to undergo longer downward migration during the early life stage. Otolith $\delta^{13}\text{C}$ profiles suggested that the fish with a longer migration distance had a higher metabolic rate in their early life-history stages than in the later stages. However, the metabolic rate did not vary for the fish living within a narrow vertical range during their larval to adult stages. In conclusion, ontogenetic vertical migration or residential contingents of deep-sea benthic fishes varied among taxonomy, reproductive style and habitat depths, displaying different adaptations or life strategies to the deep-sea environment.

OtoChuki Session (IVD, Friday)

Abstract reference: IVD_Tribuzio_01

A New Era of Ageing Spiny Dogfish (*Squalus suckleyi*) in the North Pacific Ocean

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The methods for ageing sharks (and all chondrichthyan fish) are often different from those used for teleost fish species. Sharks do not have highly calcified otoliths and in some cases, the cartilage does not calcify enough to make annuli discernible in other hard parts. This is the case for spiny dogfish

(*Squalus suckleyi*), in which historical research deemed the vertebrae unusable; thus the dorsal fin spine ageing method was developed in the 1930s and has not changed since. Dorsal fin spine ages have been validated by both oxytetracycline tag and release studies and bomb radiocarbon. Despite the prevalence of using the dorsal fin spine ageing method, there are known caveats associated with this approach. The dorsal fin spines protrude into the environment and are often broken or worn, especially in larger, older animals, necessitating an analytic method to account for lost annuli. Dorsal fin spine ages are highly uncertain due to the difficulty in reading these structures, resulting in high inter- and intra-reader variation, and the analytic method accounting for lost annuli adds another source of uncertainty to the age estimates. This study examines a new method for ageing spiny dogfish which uses histological staining techniques on vertebrae thin sections. Spine and vertebra ages generated by multiple readers were compared to estimate inter-reader precision and the variance associated with each structure. Results suggest that ages derived from the vertebrae thin sections are similar to those derived from the dorsal fin spines; however the vertebra between-reader variability is reduced and there is no need to account for lost annuli. Results presented here are part of a larger study; these data will be used to generate improved growth models with reduced uncertainty in the growth parameters, which will be used in stock assessments.

Abstract reference: IVD_Dinh_02

Age structure of the goby *Parapocryptes serperaster* in the Mekong Delta, Vietnam, based on length-frequency and otolith analyses

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Age structure of the goby *Parapocryptes serperaster* was determined by analysing length-frequency distributions and examining the sagittal otolith in fish sampled from March 2013 to February 2014 in the Mekong Delta, Vietnam. A total of 476 individuals (246 males and 239 females) were collected in the study site in wet and dry seasons. The total lengths were 107 - 221 mm for female and 121 - 230 mm for male. Otoliths from 164 female and 196 male gobies were readable, and the average percent error (IAPE) of three reading times was 4.65%. Otolith length, width and weight were determined and the use of morphometric analyses for age identification was evaluated. Morphometric data from the otoliths had a strong relationship with fish length of the two genders. The maximum age was 3.93 years for males and 3.77 years for females. The mean age estimated by reading otolith annual rings and by analysing length frequency distribution was not significantly different, suggesting that both otolith weight and morphometrical dimension can be used as a predictor for fish age identification. The most accurate age identification can be made by measuring otolith length, followed by otolith mass and otolith width.

Abstract reference: IVD_Irgens_03

Changes in size, shape and appearance of juvenile Northeast Arctic cod (*Gadus morhua*) otoliths during settling

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Northeast Arctic (NEA) cod juveniles are known to settle in the Barents Sea during autumn after months of pelagic drift from spawning grounds off northern Norway, but limited knowledge exists on the exact timing and dynamics of the process. Each year during August and September, extensive Norwegian-Russian Ecosystem surveys are conducted in the Barents Sea and 0-group fish are sampled with pelagic trawls to calculate abundance indices of commercially important fish species, including the NEA cod. This study provides indirect evidence of early settling of NEA cod during the timing of the Ecosystem surveys, which potentially can result in biased 0-group estimates obtained from the catch data. We have analysed otoliths from settled juveniles (1-group cod) sampled in February and March and were able to identify macro-structural checks (prior to first translucent annulus) in juvenile cod otoliths. These checks are likely to be associated with the settlement which introduces a remarkable change in physical environment, prey type and food availability, as the juveniles go from warm prey-rich Atlantic water masses in the upper layers down to the dark and colder bottom waters of the Barents Sea. Based on otolith growth and back-calculation, we present estimates of fish size and timing of settlement in the 2005 and 2008 cohorts. Furthermore, we will demonstrate that age-group 0 and 1 can be discriminated based on simple otolith shape parameters due to changes in lobe formation during the period of settlement in NEA cod.

Abstract reference: IVD_Piñeiro_04

Age and growth of John Dory *Zeus faber* (Linnaeus, 1758) in Atlantic Iberian waters using vertebrae and otolith increments

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The John dory (*Zeus faber*) is an important commercial species worldwide, however knowledge of its biology is limited. In this study the age and growth of *Z. faber* were determined using otolith (sagitta) and vertebrae from specimens caught in Iberian Atlantic waters. The specimens were collected in scientific trawl surveys carried out between February and October 2012 at 77 to 255 m depth. Vertebrae and otoliths of 181 individuals ranging from 4 to 60 cm total length were analyzed. The largest female measured 60 cm total length (TL) and weighted 4 kg, while the largest male measured 42 cm TL and weighted 1.250 kg only. As a result of such differences, length at age showed significant differences between sexes. Growth parameters were estimated on the basis of direct reading of otoliths and vertebrae and compared with those obtained by other authors. Vertebrae performed better than otoliths for ageing, as they offer sharper definition of rings and growth increments. This is the first study of the growth of *Z. faber* inhabiting the eastern Atlantic.

Abstract reference: IVD_Staby_05

Post-settlement growth of hake from the Norwegian coast based on otolith microstructure analysis: Evidence for counter gradient variation?

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Little is known of the life history of European hake (*Merluccius merluccius*) in the northern parts of its range - especially in contrast to the extensive literature on populations in the Mediterranean and Southern Europe. Small hake are consistently found in the fjords of Southern and south-western Norway, but it is not clear whether these are resident fish or whether these areas are simply nursery grounds for a larger migratory population. Conditions in the fjords are generally colder than the areas inhabited by juvenile hake in other areas, and if these are resident populations, then some local adaptation may be expected. Counter gradient variation (CGV) is a common phenomenon in fish with a wide latitudinal range, and is often manifested as an increased growth rate in the most northerly populations. We might expect hake to exhibit a similar pattern. Although it is notoriously difficult to study individual growth in large hake, primary increment analysis has been successfully applied to small (young of year, YOY) individuals in the Mediterranean, Bay of Biscay, and English Channel. Here we conduct a similar analysis of YOY hake from Norwegian fjords, and combine our findings with published data to test for higher individual growth at higher latitudes, as consistent with CGV. We also use these data to develop a temperature dependent otolith growth model which may be useful to interpret further population-level patterns.

Abstract reference: IVD_Jaafour_06

Colonization, hatch-dates, and growth rates of juvenile *Hyporhamphus picarti* (Actinopterygii, Hemiramphidae), in the Nador lagoon (NE Morocco)

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Coastal lagoons are among the most productive wetlands in the Mediterranean region and play an important ecological role for many fish species. The Nador lagoon, located in the northeastern part of the Moroccan Mediterranean coast, was sampled with a beach seine at six stations during different seasons, from 2012 to 2014. Among those fish species collected, the African halfbeak, *Hyporhamphus picarti*, was one collected for the first time in this lagoon. This species was found only in late summer and autumn, and all the individuals captured were juveniles, indicating that the lagoon serves as an important nursery for this species. Otoliths (lapilli) microstructures of juvenile *H. picarti* were analyzed to estimate and compare growth rates and hatch dates of individuals caught in 2012 and 2013. The total lengths of juvenile *H. picarti* were (68.10 +15.19 mm) and (101.03 +28.77 mm) in 2012 and 2013, respectively, and their estimated age varied from 39 to 58 days in 2012, and from 42 to 76 days in 2013. For both years, the relationship between fish size (TL) and age (days) was described by the following linear equations: $TL = 1.88 \times \text{age} - 19.29$ ($n = 52$; $r^2 = 0.63$; $p < 0.05$) in 2012, and $TL = 2.35 \times \text{age} - 35.57$ ($n = 29$; $r^2 = 0.71$; $p < 0.05$) in 2013. Juveniles collected in 2013 had significantly higher growth rates (somatic and otolith growth); 1.88 mm d^{-1} and 2.35 mm d^{-1} in 2012 and 2013, respectively. For both years, the average otolith increments width increased until about the 25th day (corresponding to a size of about 28 mm TL), and then

decreased. Back-calculated hatch dates of juveniles collected in the lagoon indicated that they hatched in summer from June to September. These results indicate that *H. picarti* has a short spawning period during the warm period, and use the lagoon as a nursery ground in late summer and autumn.

Abstract reference: IVD_Dufour_07

Determination of strontium chemical environment and elemental mapping of otoliths by synchrotron radiation techniques

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The record of environmental conditions by otolith chemistry makes them invaluable tools in fish ecology and fisheries research. Strontium is the most often-used element because of the quality of environmental information it provides for reconstruction of fish life history and its abundance in otoliths. The reliability of the Sr quantification depends on the mode of incorporation of the element in the aragonite lattice and its permanence over time. The random substitution of Sr for calcium in the aragonite has long been assumed and was recently proved by Doubleday *et al.* (2014) for modern fishes at the macroscale. The present study assessed whether this substitution can be considered as homogenous at the microscale and permanent. A set of modern and archaeological *Micropogonias* sp. and *Galeichthus peruvianus* otoliths from South America was analyzed by a combination of point strontium K-edge X-ray absorption spectroscopy (XAS) and comparative micro-X-ray fluorescence (μ XRF) analysis at diagnostic energies accurately selected by monochromatisation of a synchrotron beam. Our evidence supports that Sr speciation is identical independent of its concentration, location in the otolith, species studied, and individual and geological age. Otoliths provide reliable archives for reconstructing fish palaeoecology and palaeoenvironments over thousands of years. We have obtained fluctuation maps of strontium content using quantitative synchrotron μ XRF with a $4 \times 3 \mu\text{m}^2$ (H \times V) on-sample spot size, the highest lateral resolution macroscale maps known. Despite the limited availability of synchrotron beamtime, the technique appears very promising. It provides fast and precise elemental maps to assess fish life history at a very high resolution without the restrictions inherent to otolith axial elemental profiles generated by the most commonly used techniques.

Abstract reference: IVD_Sirot_08

Past and recent growth patterns in two species with contrasting demographic responses to environmental changes in a tropical lagoon

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In order to better understand the causes for inter-specific difference in fish population responses to 30 years of environmental changes in a vast tropical lagoon (the lagoon of Terminos, Mexico), past and recent growth patterns were compared between two species showing opposite demographic trends in the area: the Silver Perch *Bairdiella chrysoura* (Scianidae) experienced a strong decline in abundance while the Western Atlantic seabream *Archosargus rhomboidalis* (Sparidae) kept its population constant. About 400 otoliths, from adults and juveniles of the two species caught at three distinct time periods (1980, 1997, and 2006-2011), were used to characterize inter-specific differences in growth patterns and their temporal modifications. After validation of the time for opaque marks deposit in the study area, seasonal growth increments were used to estimate individual age (in months) for both species. These data were then used to model population growth curves and average lifetime growth patterns for each species and period. Our result showed that both species shared similar growth patterns with no significant change since the 80's. However, temporal comparisons of lifetime growth profiles revealed that, contrary to *A. rhomboidalis*, *B. chrysoura* suffered a non-negligible decrease (-15%) of its average growth rate during early juvenile life (*i.e.* during the first four months following settlement in the lagoon) over this period. This modification helps elucidating why *B. chrysoura* experienced a severe population decline in the lagoon since 1980, while in the meantime *A. rhomboidalis* proved resistant to environmental perturbations.

Abstract reference: IVD_Malca_09

Age and growth of larval Atlantic bluefin tuna, *Thunnus thynnus*, from the Gulf of Mexico

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Atlantic bluefin tuna (*Thunnus thynnus*) are managed through international agreements under the auspices of the International Commission for the Conservation of Atlantic Tuna (ICCAT). Bluefin tuna are highly pelagic, undertaking extensive migrations throughout the Atlantic, but the vast majority of spawning takes place in the Mediterranean Sea and Gulf of Mexico in April-May. Despite 30 years of ichthyoplankton surveys in the GOM there is little known about larval bluefin growth and survival. Larval daily age can be determined directly by examining otolith microstructure. In this study, we describe preliminary age-length relationships for larval Atlantic bluefin tuna using otolith microincrement analysis. Larvae were collected from plankton tows during the spring ichthyoplankton GU1201 research cruise using 1 x 2 m (S10) nets (505 micron mesh) in the Gulf of Mexico in April-May 2012. Sagittae and lapilli were dissected from 50 larvae, ranging from 2.4 to 7.4 mm (NL or SL). Estimated ages ranged from 3 days to 14 days. From these data we developed new growth curves for both the eastern and western Gulf of Mexico. Growth was highly variable at a given length, which likely reflects environmental variability encountered in the complex and dynamic oceanographic environment of the Gulf. Results will improve the annual larval index that is based on specimens collected solely off South Florida. In addition, results will be used in the Atlantic bluefin tuna stock assessment and play a key role in developing predictive recruitment models.

Abstract reference: IVD_Vasconcelos Filho_10

Shape analysis of sagittal otolith of *Haemulon plumierii* caught in Pernambuco State (Brazil): ontogenetic and population perspectives

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Haemulon plumierii is a small fish distributed from the southeast coast of the United States to the southeast Brazil. It is a carnivore species, found during the day in dense aggregations on reefs, around coral formations, or on sandy bottoms. This species represents an important economic resource both for recreational diving and artisanal fisheries in Pernambuco State (Northeast Brazil). The available information on *H. plumierii* on the studied area is restricted to fisheries, with scarce information regarding biological aspects. Therefore, since the otoliths encrypt record life history events which can be used as sources of information for ecological and demographic studies, we analyzed the shape variation of sagittal otolith of *H. plumierii* during its life and along the fishing area, aiming at supplying the first information on the ontogeny and population structure of this species based on shape descriptors of the sagittal otolith contour. Using a sample of 123 individuals (both immature and mature), we found significant linear relationships ($P < 0.05$) between fish length and several otolith shape indices (area, perimeter and maximal and minimal Feret diameters), and a non linear relationship between fish size and otolith weight. Otolith circularity was not significant. All significant variables presented negative allometric when related to fish length, with no difference between males and females, or left and right otoliths. To avoid influence of fish size, we investigated the population structure throughout the fishing area (North, Center and South of Pernambuco) based on 53 mature individuals between 20 to 25 cm (TL). Regarding the phenotype variation, principal Component Analysis (PCA) of the first 20 harmonic amplitudes suggests only one population of *H. plumierii* in Pernambuco. Thus, one expects the same growth rates for the entire area, with no differences between sexes.

Abstract reference: IVD_Elking_11

Larval Otolith Formation in Striped Bass (*Morone saxatilis*)

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Otolith (earbone) studies have become more prevalent in recent years as use has expanded from only aging to examination of migration and fidelity to natal habitats. The otolith of striped bass, *Morone saxatilis*, is used for all of these purposes, yet its formation and early development have not been documented. We identified the timing and formation of the three otolith pairs during late pre-hatch embryo, yolk-sac larvae, and post yolk-sac larval stages using a dissection scope and measured both fish and sagittal otolith length. The sagittal otoliths were first to appear, forming shortly before hatch and were observed growing larger throughout the larval stage. The lapilli otoliths formed within the first 24 hours post hatch. The asterisci otoliths were more difficult to locate but formed between 5 and 15 days post hatch. The larval sagittal otolith growth can be

divided into two stages. Stage 1 is when the otolith is spherical and grows in a quadratic relationship relative to fish total length during yolk-sac development (5 ph). Stage 2 starts by about 15 dph and is characterized by the otolith elongating along the anterior/posterior axis and the sulcus forming. This knowledge of the when the otoliths form will affect any microchemical analysis done on the first year of life since all three otoliths form at different times. For example, if natal habitat is being studied then the sagittal should be analyzed at it forms prior to hatch while the asteriscus does not form until after the yolk-sac stage.

Abstract reference: IVD_Sturrock_12

Using otolith strontium isotopes to reconstruct life history portfolios within salmon populations: When do different phenotypes contribute?

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The maintenance of life history diversity within and among populations is thought to be critical for the persistence of salmon stocks. Preserving and restoring diversity in life history traits is thus central to many recovery efforts, but it is necessary to first understand how environmental factors affect their expression and success. Rotary-screw trap sampling in the California Central Valley indicated that juvenile salmon outmigrate at different sizes, ages and times of the year. We used otolith strontium isotopes in adult Chinook salmon returning to the Stanislaus River, a regulated stream at the southern end of the species range, to determine the influence of river conditions on juvenile behavior and survival. Paired otolith and scale samples were used to reconstruct juvenile outmigration patterns of successful adults from cohorts emigrating between 1999 and 2011. For each adult, the size it had outmigrated from its natal tributary was reconstructed by coupling otolith strontium isotope and radius measurements. Size distributions and phenotype contributions were compared between the juvenile outmigrants and adult “survivors”, and used to identify trends in selective mortality. Our seven focus years exhibited contrasting flow regimes as a result of differences in precipitation patterns and local water operations. In wetter years, the majority of juveniles outmigrated as fry, while in drier years, outmigrants tended to be fewer but larger. Metrics of outmigration behavior (abundance, size and phenology) varied primarily as a function of hydroclimatic regime, while survival rates appeared to be driven more by conditions within the natal tributary, and size- and time-selective mortality. While fry survival is generally assumed to be negligible in this system, our data indicate that they can represent more than 20% of the reproductive population. Patterns in juvenile outmigration behavior and survival are discussed in the context of water and fisheries management, and the portfolio effect.

Abstract reference: IVD_Kuroki_14

Contribution of otolith microchemistry to reveal the evolutionary origin of catadromous

migration in anguilliformes fishes: an example of tropical moray eel

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Freshwater eels of the anguillidae are generally believed to be only one family with diadromous life history in anguilliform, while other 18 families are considered to be strictly marine species with some exceptions such as a few species of the Muraenidae and Ophichthidae that have occasionally been found in freshwater or estuaries. However, why and how the freshwater eels can invade into the freshwater habitat and start their catadromous migration is unknown, and is a special interest for scientists. Here we report a hint to speculate the evolutionary origin of catadromy in freshwater eels by analyzing the otolith microchemistry of a Muraenidae that shows an exceptional migratory history in anguilliform fishes. A moray eel *Gymnothorax polyuranodon* is one species that is known to be present in freshwater in the Indo-Pacific, but its life history is unknown. To evaluate the patterns of freshwater use by this species of marine eel, the otolith Sr/Ca ratios of four *G. polyuranodon* (275–344 mm) caught in a freshwater stream of Fiji were analyzed. The consistently low Sr/Ca values (0–4) indicated upstream movement after settlement and freshwater or estuarine residence of all four individuals. These eels did not appear to have entered seawater just for a short time period, suggesting that *G. polyuranodon* might be a catadromous species of marine eel. Recent discovery of a 'living fossil' eel (Anguilliformes: Protoanguillidae) from an undersea cave in Palau clearly showed the marine origin of entire anguilliformes. Furthermore, molecular phylogenetic analysis of all 19 anguilliform families showed that the anguillids originated from mid water anguilliformes fishes such as nemichthyidae, serrivomeridae, saccopharyngidae and eurypharyngidae. All these considerations will support an idea that the freshwater invasion of this *G. polyuranodon* of Muraenidae demonstrates the early stage of evolutionary process of catadromy in freshwater eels of anguillidae.

Abstract reference: IVD_Moreira_15

Use of stable isotopes (oxygen and carbon) in otoliths to study fish movement and connectivity in *Lipophrys pholis*

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The shanny *Lipophrys pholis* is a common intertidal fish found in Portuguese coastal waters. Spawning takes place from early winter to late spring in rocky beaches, after which demersal eggs hatch and larvae disperse along the coast. About two months later, young fishes (juveniles?) return to the beach and settle in a particular set of tide pools, suggesting a homing behaviour. Information

on coastal recruitment and connectivity processes are, however, scarce for this species. Stable isotopes recorded in fish otoliths may provide new insights about the home residency, movements, structure and connectivity of fish populations. Therefore, in this study, stable carbon ($\delta^{13}\text{C}$) and oxygen ($\delta^{18}\text{O}$) isotopes in *L. pholis* otoliths and surrounding water were investigated. For that, 120 juvenile fish (16 to 26 mm) were collected during May 2014 in six rocky beaches (Agudela, cabo do Mundo, Boa Nova – three sites within one region; Peniche, Sines and Olhos de Água – three additional regions) along the Portuguese coast. The obtained regional and inter-site data was overlapped with historically available sea water surface temperatures. New findings are discussed in light of the *L. pholis* life cycle knowledge.

Abstract reference: IVD_Vitale_16

Age estimation and otolith analysis of *Fistularia commersonii* Rüppell, 1838 (Syngnathiformes – *Fistularia*) in the central Mediterranean Sea

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Otoliths features of the Indo-Pacific Lessepsian species, *Fistularia commersonii* Rüppell, 1838 has been investigated through the specimens occasionally caught by commercial fleet off the Southern coasts of Sicily. Investigations on morphometric, meristic and age (based on otoliths analysis) were performed using a total of 23 specimens. The total lengths of the collected sample ranged between 82.5 and 119.5 mm with a sex ratio (F/sexed specimens) of 0.74 and with a gonad investments up to 3% of body weight. An accurate analysis has been performed on the otolith size, in view to identify the successive distinguishable zones (“age related”) from checks “ontogenic” or “life history” crucial moments such as hatchling, settlement or spawning. The age-length key, with a maximum estimated age of 5 years old, represents the first attempt to recognize the ages of this species in the central Mediterranean sea based on otolith interpretation.

Abstract reference: IVD_Hüssy_17

Otolith microchemistry: A useful tool for age validation?

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In the Baltic Sea two cod stocks are found, the “Eastern Baltic cod” and “Western Baltic cod”. One of the most persistent problems in relation to stock assessment of the Eastern cod stock is an acceptable level of precision in age estimation. Owing to the hydrographical regime in the Baltic Sea coupled with the migration dynamics of the cod, the otolith microstructure is characterised by low contrast between growth zones and irregular zone formation. Age estimation of the Western stock on the other hand is considered highly precise owing to the high contrast between growth zones. The objective of this study is to explore the applicability of otolith microchemistry patterns from the nucleus to the edge of the otolith for age validation. Otoliths of 4 year old individuals (agreed age

reading from three independent readings) were selected and profiles of opacity and trace elements standardised to a common length. Patterns of opacity and trace elements from the Western stock were used to identify elements with signatures matching growth zones with respect to maxima and minima at the same distances from the nucleus. Eastern otoliths from were then examined for the occurrence of similar patterns in otolith trace elements. Matching patterns with respect to occurrence of minima and maxima in both otolith opacity and microchemistry of Western cod were found for Mg and Rb, Ba and Cu (to less extent Zn and Zr). No match was found for Pb, Sr, Mn, Na, Li. In Eastern cod however, only Mg and Rb showed a periodical pattern with maxima and minima at the same distances from the nucleus as the Western stock. We therefore propose that the incorporation of these two trace elements into the otolith may be linked to fish somatic growth and thus presents a tool for age validation in species where ageing is difficult.

Abstract reference: IVD_Neil_18

Can we derive a deep water $\delta^{14}\text{C}$ curve to aid age validation of NZ deep sea fish species?

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Determining the correct age of fishes, and then having the ability to calculate their growth rate, their mortality rate, and the age structure of exploited populations, is a fundamental requirement in fisheries science. Longevity, in particular, is an important measure of a species' productivity. Several of New Zealand's deep sea species (bluenose - *Hyperoglyphe antartctica*, rubyfish - *Plagiogeneion rubiginosum*, black oreo - *Alloctytus niger*, smooth oreo - *Pseudocyttus maculatus*, black cardinalfish - *Epigonus telescopus*) have had the bomb chronometer method of radiocarbon ageing used to validate the relationship between ages determined by the standard procedure of counting growth zones in otoliths, with ages determined by the bomb chronometer procedure of radiocarbon dating. The juveniles of many of these species reside near the surface, and so, if aged correctly, their otolith core $\delta^{14}\text{C}$ should confirm with curves derived from surface waters (e.g. *Pagrus auratus*). However, not all species have a juvenile phase that reside within well mixed surface waters. Here we attempt to derive a deepwater $\delta^{14}\text{C}$ curve from otolith edge samples (samples that represent the adult deep dwelling phase of deep sea species) that can be used to aid validation in deep dwelling species. These otolith edge samples were found to fall consistently below and to the right of surface water bomb-carbon curves, indicating a 10-15 year lag in ^{14}C uptake.

Poster Session (IVE)

Abstract reference: IVE_Sainza_01

Age and Growth of European conger eel (*Conger conger*) in North East Atlantic Ocean using otoliths and vertebrae.

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A sample of 1107 fish served to age and to analyze growth of European conger eel (*Conger conger*) from NE Atlantic Ocean using otoliths and vertebrae. Sizes sampled were comprised between 21 and 156 (TL, cm) with most specimens within 33 and 55 cm. The largest individuals were females and males were smaller than 71 cm. Length-weight relationship showed an allometrical growth. Females were heavier than males. Length-weight relationship parameters were: $a = 0.00005$, $b = 3.81$, for females and $a = 0.0004$ and $b = 3.33$ for males. Females were larger and live longer than males; the maximum estimated age was 17 years for females and 7 years for males. The comparison of mean ages obtained using both calcified structures were not significantly different, even if the growth pattern was clearer in vertebrae than in otoliths. Vertebrae seem to be the best structure for ageing European conger eel, since they had a better ring definition and resulted in more adequate growth patterns. Length-at-age values showed differences between sexes. Von Bertalanffy growth parameters by sex and their uncertainty were calculated applying a non-parametric bootstrap method. For females, the von Bertalanffy population growth curve estimates were $L_{\infty} = 300$ cm, $k = 0.04$, $t_0 = -1.3$. For males, these estimates were $L_{\infty} = 100$ cm, $k = 0.07$ and $t_0 = -5.6$. To explore the growth at larvae stage, four otoliths of pre-metamorphic larvae were analyzed and the results showed a mean increment width of $0.81 \pm 0.06 \mu\text{m}\cdot\text{day}^{-1}$ for individuals ranged between 12.8 and 13.6 (TL, cm). All the results are compared with those obtained by other authors.

Abstract reference: IVE_Matić-Skoko_03

Age determination and validation on otoliths of the striped red mullet, *Mullus surmuletus* from the Adriatic Sea

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There is a need to establish a method for age determination of the striped red mullet, *Mullus surmuletus* as target fish species of Mediterranean artisanal fisheries. In total, 387 specimens ranging in total length from 6.8 to 32.9 cm were analyzed for this purpose. Ground otoliths were used to estimate fish ages. The annual periodicity of annulus deposition was supported by the marginal increment analysis. The annual growth of otoliths was verified using edge-type analysis.

The formation of growth increments followed a seasonal pattern. The proportion of otoliths with opaque margins was the highest (>80%) in the winter period (December-March). Moreover, the monthly mean marginal increment showed a single minimum in June and confirmed the formation of one annulus per year. A distance measurements analysis revealed that first regular ring appears at distance of 0.78 mm (± 0.104 SD) from otolith nucleus and fish age of about 0.83 years. The age analysis revealed 6 age classes. Predominance of age classes 1+ and 2+ in the total catch (54.3% individuals) was observed. All measured otolith morphometric parameters (length, width and mass) were linear with fish age. The linear model explained between 80.3% and 84.4% of the variation in age. The most precise age estimations of analyzed population were obtained from the otolith width, followed by the otolith length and mass. The growth parameters were estimated by the fit of the Von Bertalanffy growth function. The growth of *M. surmuletus* was rapid up to 2 years of age and then slowed. Considering the identified biological implications, existing management should be complemented by a more holistic, ecosystem approach to regulations, in order to assure effective conservation measures for this economically and ecologically important fish species.

Abstract reference: IVE_Vittori_04

Age estimation and back-calculation of fish length of the European hake, *Merluccius merluccius* (Linnaeus, 1758) in Sardinian waters.

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Aim of the present study is to evaluate the growth parameters for the European hake, *Merluccius merluccius*, in Sardinian waters by means of otolith annual ring analysis, providing also an indirect validation through the use of back-calculation. A total of 283 individuals (of which 150 males, ranging from 8.5 to 42.1 cm TL, and 133 females, that ranged from 11.2 to 83.3 cm TL), collected during the MEDITS survey in 2012 and 2013, were selected for the age reading. The determination of the age was carried out on the whole otolith for fish with total length smaller than 20 cm while for the others a transversal section across the core was prepared. Hyaline rings count was performed twice by two different readers, showing good reproducibility (%CV= 13.9; IAPE= 10.69). The estimated von Bertalanffy parameters (\pm s.e.) were $\text{Lin}f = 53.26 \pm 10.15$, $k = 0.17 \pm 0.06$, $t_0 = -1.73 \pm 0.26$ and $\text{Lin}f = 102.77 \pm 13.67$, $k = 0.09 \pm 0.02$, $t_0 = -1.46 \pm 0.13$ respectively for males and females. Back-calculation, carried out on otolith sections through the measurement of the ventral radius of each annulus, has provided an estimation of fish size at each year. Mean length at each age class (1-6 for females, 1-4 for males) obtained with the two methods was compared individually showing no statistically significant differences (t-Student, p-value > 0.05), except for age class 1 for both sexes. This result is probably due to the difficulty, well-known in literature for the European hake, in the identification of the first year annulus, which could be confused with one of the false rings around the nucleus. Nevertheless, this procedure certainly offers a valuable mean for corroboration of the age determination when other validation analysis are not achievable.

Abstract reference: IVE_Jean-Christophe_05

**Age estimation of the brown trout, *Salmo trutta* L introduced in the Kerguelen Islands:
Determination of the otolith optimal zone for annuli detection.**

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In the Kerguelen island (49°S, 70°E) little knowledge exists on the growth of salmonids otolith and the annuli formation. At this latitude, only few studies used otoliths as a tool to estimate brown trout ages. The validation of age estimation method and the characterisation of an optimal zone where to make measurement and laser ablation (LA-ICPMS) appear as a fundamental preliminary stage before developing population study. The aim of this study was to 1) define a zone of the sagittae which optimize the probability to make reliable and repeatable measurements and which also minimize the inter-individual variability of these measurements. This zone will help to select the axis where LA-ICPMS transect have the maximum probability to cross annuli. 2) To validate the position of the first annulus, the annual formation of subsequent annuli, and characterize the variation of each annuli location. After Alizarin Red S otolith marking and 3 years of fish recapture 53 sagittae were studied and compared with scales. A composite index was created to select an optimal zone. Results show that the area between 80° and 120° compared to the rostrum/core axis was optimal to measure distances. Brown trout mark seasonal annulus as of the first winter after hatching. The location of annuli was well identified up to five years. In the case of Kerguelen fish, straight relationship exists between fish size and otolith radius. This relation validates otolith as a reliable tool for brown trout length-at-age back calculation. The actual temporal resolution of the elemental composition through LA-ICPMS transect depends on both the sampling resolution of the laser ablation and the otolith growth rate. In this context, the proposed formal approach allows distinguishing area of interest that consists in the best compromise between spatial resolution (main growth axis) and precise age positioning.

Abstract reference: IVE_Ventero_06

Anchovy (*Engraulis encrasicolus*) otoliths reveal growth differences between two areas of the Spanish Mediterranean Sea

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In the Mediterranean, small pelagic provide the larger catches, based mainly on 2 species: anchovy and sardine (*Sardina pilchardus*). Although the Mediterranean Sea is considered oligotrophic, there are areas with increased production due to their unique conditions related to the bathymetry, hydrography and meteorology. In the Spanish Mediterranean Sea, there are two areas with increased production due to their environmental characteristics: the Ebro Delta continental shelf, due to the discharge of the Ebro River and the Alborán Sea that receives Atlantic water, rich in nutrients, through the Strait of Gibraltar. The amount of growth in the first year can be measured

from the otolith (R1) and some cases may be suitable to use it as a tool for stocks separation. This study presents and analyses anchovy otolith R1 distributions from these two different areas, from samples collected during acoustic surveys in the Spanish Mediterranean in 2012 and 2013 during the months of June and July. In addition, the condition factor was estimated for the total number of individuals and for the sexes separately. Regression analysis, similarity analysis, principal components analysis (PCA) and the Wilcoxon-Mann Whitney test were undertaken in order to identify any different growth patterns. The results suggested that the differences may be characteristic of the existence of population groups, and these could be used as a tool for differentiating stocks.

Abstract reference: IVE_Ferri_07

Assessing the use of otolith morphometrics to predict the age of the black scorpionfish, *Scorpaena porcus* (Linnaeus, 1758)

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The otoliths of the scorpaenids are relatively large and therefore easy to handle for the measurement of age, but numerous ramifications at the otolith edges can make the process of annual growth zones counting complex. One of the alternatives to this subjective, expensive and time consuming technique is the use of otolith morphometric parameters to derive the age of fish. To test the practicality and accuracy of the use of otolith morphometrics (length, width, thickness and mass) to estimate the scorpaenid age, we examined sagittal otoliths of 510 specimens of the black scorpionfish *Scorpaena porcus*, collected from the eastern Adriatic. The inner structure of the ground otoliths of *S. porcus* displayed an alternating pattern of translucent and opaque zones around an opaque nucleus. The annuli in the otoliths are laid down annually, the opaque zone being deposited in spring-summer and translucent zone in autumn-winter. Observed maximum ages for males and females were 10 and 12 years, respectively, although most of the sampled fish were 4 and 5 years old. The power model described the relationship between otolith morphometrics and age, explaining between 19.4% and 53.4% of the variation in age. The most precise age estimations were obtained from the otolith mass, followed by the otolith length, width and thickness. Relationship of the observed age and age estimated from all measured otolith morphometrics were described by a simple linear regression, accounting for 32.1% - 60.5% of the variation in estimated age. However, 95% confidence intervals did not include intercept =0 and slope =1 for all equations. Although the use of analysed otolith dimensions saved time and provided a similar population age structure to that of grounded otolith ages, this alternative method is not an accurate estimator of *S. porcus* age, primarily due to the length overlap between the different age classes.

Abstract reference: IVE_Carvalho_10

Using otolith microchemistry as a tool on migration studies of *Anchoa tricolor* (Spix & Agassiz, 1829) along its ontogenetic development in an subtropical estuary at South America

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The main goal of this work is the description of *A. tricolor* migrations along its ontogenetic development through Sr:Ca and Ba:Ca ratios of sagitta otolith using microchemistry. There were made monthly sampling on spring-2010, summer-2011 and autumn-2011 in three sectors of Paranaguá Estuarine Complex (ECP) (internal, intermediate and external), Brazil (25° 15' -35' S e 48° 20' - 45' O). These sites were also characterized on its salinity and pluvial precipitation. There were analysed 198 otoliths of *A. tricolor*, with the length ranges of 25-35 mm (n=33), 36-45 mm (n=22), 46-55 mm (n= 28), 56-65 mm (n= 45) e 66-75 mm (n= 70). The otoliths were cleaned with MilliQ water, digested in nitric acid (HNO₃) 10% and the Sr, Ba e Ca concentrations were determined on ICP-OES. The results showed variations on Sr:Ca and Ba:Ca ratios for the different sectors of the estuary on the different fishes' length classes. The lowest mean for the Sr:Ca ration was found on the length interval between 36-45mm, on the internal sector during summer (3,39 DP =± 1,39); on the other hand the highest mean occurred at the 46-55m interval for the external sector also during summer (11,53 DP = ± 2,84). The highest mean for Ba:Ca ratio (0,23 DP = ± 0,021) was detected on the 25-35mm class interval for the external sector during summer and the lowest mean of this ratio occurred during autumn at the internal sector for the animals between 66-75mm (0,012 DP= ± 0,0007). These values indicate that *A. tricolor* has an environmental plasticity inside CEP along its ontogenetic development. The seasonality of the rain on the area increases the influence of the Ba:Ca ratio, since during the period with higher pluvial precipitation (summer) there's more barium available in the environment due to higher fluxes of freshwater in estuary.

Abstract reference: IVE_Piñeiro _12

Seasonal climatic conditions influencing daily growth pattern on young of the year European hake (*Merluccius merluccius*) off NW Spanish waters

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Daily growth of Atlantic juvenile hake (*Merluccius merluccius*) from Northwest Spanish Iberian coasts was estimated by counting daily increments of the transversal sections of otoliths. The microstructural analysis of otoliths enabled to estimate seasonal variability of growth rates. Juveniles were sampled in one of the most important recruitment areas of the species of NW

Atlantic Iberia in surveys that were carried out during spring and summer 2012. A total of 103 otoliths (sagittae) of individuals ranging from 1 and 25 cm total length were analyzed. The results indicated that juvenile growth show growth pulses of varying intensity throughout the year related with climatic conditions. The evolution of daily increments widths indicate higher growth rates occurring in spring-spawned juveniles in comparison with the winter-spawned specimens, clearly manifest by the increase of the average daily increments widths deposited in otoliths. Hake juveniles have shown growth rate increase during spring in apparent synchrony with the seasonal upwelling regime off NW Galician waters. This study was made possible by the financial support of project CRAMER-MICINN and ECOPREGA-Xunta de Galicia.

Abstract reference: IVE_Nuñez_15

Determination of larval growth stages in the codlet *Bregmaceros cantori* (Pisces: Bregmacerotidae) in Cariaco Trench, Venezuela

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The striped codlet *Bregmaceros*, only genus of Bregmacerotidae family, are small fish living in deep and anoxic waters of Cariaco Trench at eastern Venezuela. Otolith zones were described in daily growth rings on larval and juvenile sagittae otoliths of *Bregmaceros cantori*. Samples were taken with Bongo and microneckton nets, from February to April 2013. Since the hatching mark, the presence of four differentiated zones on the otolith was observed: pre-hatching (10 µm core diameter), pre-fixation (18 rings of 0.5-6 µm wide), a middle one with wider increments, and a last one with regular and transparent rings (10-15 µm). Rings are wider between 18 and 41 days of age, matching with the change from pro-larvae to post-larvae. According to our age estimations, stages could be separated in: planktonic larvae (18 days), post-larvae (37 days) and juvenile (>41 días). A faster growing was found during rainy season (0.30 mm/day) compared with the observed during dry season (0.043 mm/day). This may be explained for the water turbulence caused by trade winds, which could make difficult the feeding process for this species during that season in particular. We conclude that the marked change in the wide of daily growth rings on sagittae otoliths, might be use as evidence of growth stages duration in the adaption of this species to extreme environments such as the ones in anoxic water (depths >200 m).

Abstract reference: IVE_Vaz-dos-Santos_16

Do otoliths express the polyphasic growth of the main species in a coastal subtropical ecosystem?

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Araçá Bay is a complex coastal subtropical ecosystem (23°48'47,3"S 45°24'22,1"W) of Brazilian

coast, constituted by different marine environments and subject to a lot of human impact. The Bay was selected for an ecosystemic study and its ichthyofauna was sampled seasonally (Oct-2012 to Jan-2014) employing eight different fishing gears. The most abundant species were *Eucinostomus argenteus* (23<Lt<145mm), *Atherinella brasiliensis* (38<Lt<129mm), *Haemulopsis corvinaeformis* (68<Lt<176mm), *Mugil curema* (61<Lt<389mm) and *Diapterus rhombeus* (73<Lt<235mm). In order to analyze the fish growth, sagittal otoliths were removed and measured (Lo, mm). Taking into consideration the hypothesis that the allometric constant b of the Huxley model ($y=axb$) may change throughout the fish development and a model with stanzas is necessary, power functions were fitted between total (Lt, mm) and otolith (Lo, mm) length. The nonlinear iterative least squares method was applied and the models obtained were verified through residual analysis and the tendency of estimated b' values ($b'=\log[Lo/a]/\log Lt$) for each size class plotted against total length. If tendencies were detected, polynomial models were adjusted. Results showed that the Huxley's model was suitable for the five species. Residual analyses revealed no trends and graphs of Lt versus b' were linear, evidencing that the allometric constant does not change during the ontogeny of these species. Constant values obtained were: $a=0.126$ and $b=0.72$ for *E. argenteus* ($n=162$), $a=0.050$ and $b=0.88$ for *A. brasiliensis* ($n=92$), $a=0.188$ and $b=0.73$ for *H. corvinaeformis* ($n=122$), $a=0.056$ and $b=0.93$ for *D. rhombeus* ($n=125$) and $a=0.233$ and $b=0.62$ for *M. curema* ($n=137$). These results show that eventual changes in the growth rate do not affect the otolith development: Lo was not sensible enough to reveal the polyphasic growth, although indirect and direct growth analyses indicated the opposite. These results will be compared for a better understanding of the use of Araçá Bay by these species.

Abstract reference: IVE_Cisterna_17

Estimation of the age of jack mackerel (*Trachurus murphyi*) using daily growth rings in sagittae otoliths

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Growth of Chilean jack mackerel (*Trachurus murphyi*) was estimated by reading daily increments in sagittae otoliths of 187 individuals from 4 to 60 cm fork length (FL), collected in south-central of Chile in the period 2004-2011. From reading of daily increments was performed an adjustment between the density of daily increments and otolith radius ($n=5787$). With the integration of the ratio of otolith was estimated the individual age (in days), which was then transformed to years. Asymptotic length ($(L_\infty)=75$ cm FL; growth coefficient ($K)=0.16$ yr⁻¹; theoretical age at which length is zero ($t_0)=-0.13$ years. This research addresses two complementary methods used to estimate the age of fish. First, was analyzed annual growth rings of individuals between 18 to 60 cm FL ($n=7610$) in the period 2004 to 2011. Asymptotic length ($(L_\infty)=74.7$ cm FL; growth coefficient ($K)=0.1$ yr⁻¹; theoretical age at which length is zero ($t_0)=-0.5$ years. The second method was to use otolith weight as a predictor of fish age in years ($n=75$), in the period 2004 to 2011. Asymptotic length ($(L_\infty)=75$ cm FL; growth coefficient ($K)=0.15$ yr⁻¹; theoretical age at which length is zero ($t_0)=-0.48$ years. Growth rates of jack mackerel obtained by the methods of weight otolith and daily increments are not statistically different, suggesting that the weight of the otolith can be used as a good predictor of age of jack mackerel. The results obtained were consistent with those described for jack mackerel in Peru.

Abstract reference: IVE_Frothingham_18

Exploring Arctic Cod *Boreogadus saida* life history from nearshore Beaufort Sea with the use of otolith microchemistry

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Climate change and the recent oil and gas exploration in the arctic requires the understanding of arctic ecosystems to better recognize the implications to the region. Arctic Cod *Boreogadus saida* play an important role in the arctic ecosystem due to their abundance and ability to adapt to a wide range in habitat. Limited information exists for arctic cod life histories in the Beaufort Sea. Recent studies by the University of Alaska Fairbanks indicate the use of otolith microchemistry, specifically the ratio of barium to calcium, has the potential to be a useful natural chemical tag to help answer questions about fish movement, spawning location and timing, and stock identification in arctic cod. Archived Arctic Cod otoliths collected from nearshore Beaufort Sea (Simpson Cove n=25, Jago Lagoon n=29) in 1988 and 1989 by the US Fish and Wildlife were analyzed with the use of laser ablation inductively coupled plasma mass spectrometer (LA-ICPMS). Ratios of trace elements such as barium, magnesium, zinc, strontium to calcium were measured and examined across the length of the otolith. Multiple peaks of barium were present in many Arctic Cod otoliths indicating possible preference for riverine habitat. Potential trends in trace elements reflect the water chemistry and preferred habitat of an abundant and vital species such as the Arctic Cod and is therefore imperative as changes continue in the arctic.

Abstract reference: IVE_Katayama_19

Four types of otolith opaque zone

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Alternative formations of opaque and translucent zones in the otolith are widely used for age determination. However, the mechanisms causing annual periodicities in these zone formations is unknown, so there have been various interpretations and explanations concerning the appearance of opaque zones. In the previous symposium, our group reported two types of opaque zone with quite different structures. Further studies were performed to understand the characteristics of these opaque zones, using otoliths from various fishes. It was found that the structural heterogeneity can actually be classified into four (rather than two) types of opaque zone: **Type A:** A dark opaque zone, displaying minute, dense crystals, typically formed at younger ages. **Type B:** A washy black zone, which has the appearance of being smeared with ink, formed during seasons of active growth. **Type C:** An aggregation of grooves and discontinuous crystals formed during growth-stagnant periods. **Type D:** A deep groove, appearing luminous in transmitted light in etched otolith sections, formed during the spawning season.

Otolith zones of types A and B are typical in younger fish and during the growing season, apparently complementary to types C and D. The latter are formed during periods of stagnant growth and/or spawning seasons and are more typical of older fish, in which type A is absent. To be certain of precise age determination, it is necessary to understand the structural and biological characteristics producing these four distinct types of opaque zone.

Abstract reference: IVE_Bernal_20

Growth patterns of the dominant lanternfish *Ceratoscopelus maderensis* (Pisces: Myctophidae) from the western Mediterranean

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The growth and age of the lanternfish *Ceratoscopelus maderensis* from the western Mediterranean Sea (December 2009) was estimated throughout its entire lifespan, using otoliths (sagitta) of 59 individuals, from 3.5 mm (larva) to 64 mm (mature adult) standard length (SL). Sagittal otoliths were analyzed by means of light microscope in larvae. The examination of several sagitta from transforming and juvenile-adult stages was also accomplished by means of scanning electron microscopy (SEM) to determine otolith microstructure in those zones that were under the light microscopic resolution threshold. Serial photographs of each sagitta were mounted to count increment rings over the entire development of the specimens, and subsequently, estimate individual ages, under the assumption that the microincrements are formed by daily deposition (proven in other studies on myctophids). Maximum radius followed a significantly positive linear regression with body length. Three characteristic zones were identified in the otoliths apart from the central core: larval, transitional and post-metamorphic zones. The central core and hatched ring were determined on the basis of 30 larvae. In larvae ≤ 12 mm notochord length (NL), the microincrements ranged between 0.8 and 1.4 μm , accordingly with between-rings widths reported for lanternfishes from other regions. Distances between presumably daily rings increased progressively across larval development. A variable and characteristic band pattern was found in transforming (>16 mm SL) and adult stages, reflecting the ontogenetic shift experienced during the metamorphosis, coinciding with a habitat change from the epipelagic zone to deeper waters and the onset of diel vertical migrations. The transitional zone around the central core was rather variable and dim among individuals. In the largest adults, "daily-based rings" decreased towards the otolith edge with frequent changes in the deposition rate. The number of microincrements ranged from 7 to 332 in the analysed individuals, and it has been adjusted by von Bertalanffy's model as a function of the NL/SL ($r^2=0.955$, $IC=95\%$). Growth patterns deduced from otolith microstructure observations coincided with those reported for the same species in North Atlantic waters (Linkowski et al. 1992). Despite there were non-significant differences between the two regions, growth patterns suggested faster growth in the Mediterranean, where the species may reach 60 mm SL in 350 days, whilst in the Atlantic individuals may reach the same length in 400 days. A likely explanation for "faster growth" in the Mediterranean could be due to its warmer temperature regime with respect the Atlantic region..

Abstract reference: IVE_Valero-Rodriguez_21

Identifying seasonal patterns on daily growth of meagre otoliths

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Small variations of feeding input and environmental variables (e.g., temperature) are expected to produce between-day differences in somatic growth, which in turn may produce differences in the width between consecutive daily growth marks (DGMs) at the otolith. The potential relationship between seasonality and daily growth increment was studied on meagre (*Argyrosomus regius*). After polishing transversal cuts of sagittal otoliths, two independent reading sessions were made for each individual. Position of consecutive growth marks (measured with AVT Smartview and Photoshop software) were used for estimate growth increment across the reading radius. However, the identification and measurement of DGMs involve several sources of uncertainty. A failure to identify one or more daily increments (skipping) implies the overestimation of the average growth rate and the underestimation of age. Therefore, the raw data was first submitted to an alignment algorithm which actually observed DGM in one reading session and in the other session, more plausibly corresponding to the same growth mark. After alignment, log-transformed GI was modelled as linear function of age (days) and sea-water temperature. In spite that unexplained between-day variability is relatively large, after applying a standard model selection procedure, both age and temperature seems to have a relevant effect on GI. As expected, 1) the higher the temperature, the wider the GI is, and 2) GI tend to be smaller when fish is older. The resulting pattern combines a sinusoidal-like pattern related to temperature over the age-related pattern. This result demonstrates the potential usefulness of daily growth for testing the putative added effects of different variables or treatments, different from age itself. Some potential usefulness are suggested but further empirical and theoretical work (*i.e.*, providing the links between this empirical model and currently accepted growth models) are needed after this technique could be considered as a fisheries management tool.

Abstract reference: IVE_Maneja_22

Influence of temperature and fish size on otolith growth of laboratory-reared Atlantic cod (*Gadus morhua* L.) juveniles

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Otolith growth has the potential to reflect individual life history of and can provide information on corresponding environmental conditions experienced by individual fish. In this study, juvenile

Atlantic cod *Gadus morhua* L. were reared in the laboratory until 176 days post hatch (dph). The fish were held under different water temperature profiles simulating different settling histories and repeatedly marked with alizarin over a 3 month period examine the effects of temperature and fish size on otolith growth. Results showed that water temperature and fish size significantly affected the daily otolith increment of the sagitta (DOI) in both dorsal and distal otolith directions. The DOI of fish experiencing a 6°C increase or 6°C decrease in temperature over a 30 day interval were significantly different than those kept at 10°C throughout the same period. DOI of fish kept at constant temperature at 10°C was also higher in intermediate size fish (55 and 77 mm), suggesting an ontogenetic effect on otolith growth. The highest mean DOI (10.7 $\mu\text{m}\cdot\text{day}^{-1}$ dorsal; 7.4 $\mu\text{m}\cdot\text{day}^{-1}$ distal) was obtained from the group with temperatures increasing from 10 to 16°C, while the lowest mean DOI (3.4 $\mu\text{m}\cdot\text{day}^{-1}$ dorsal; 1.9 $\mu\text{m}\cdot\text{day}^{-1}$ distal) was observed in the group kept at 4°C. This study showed that otolith growth patterns have the potential to reflect ambient temperature history of juvenile fish during the settling stage and could be used to elucidate the cod early life history.

Abstract reference: IVE_Costa _23

Intraspecific variation in the shape of the asteriscus otoliths of *Prochilodus nigricans* in three rivers of the Amazon Basin, Brazil

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The ontogenetic allometry has been considered as a significant factor in determining the shape of the otoliths. Thus, understanding and characterization of otolith shape variation during ontogeny is of interest, since it can act as an important factor in discriminating between individuals. The aim of this study was to analyze the differences in shapes of otoliths asteriscus of *Prochilodus nigricans* to identify ontogenetic variation between individuals of different geographic areas. The curimatã, *Prochilodus nigricans* is a migratory species occurring in the Amazon River and its main tributaries. Its principal habitat is the Amazon River, its white water tributaries and its varzeas. It is one of the most popular fishes and of great economic importance in various fish markets of the Amazonian region. The otoliths of specimens were collected from commercial catches in 1996 and 1997 in sub-basins Japurá, Negro and the Amazon Basin. Based on otolith images, the shape descriptors based on harmonic were used to detect ontogenetic being individuals between locations. Contour analysis was performed based on Fourier descriptors elliptical through multivariate statistical procedures. The shape analysis revealed no significant spatial differences, but the intraspecific variation is due to the rostrum, excisure and thickness of the otolith. The intraspecific variation in the size of the otolith can be linked to ontogenetic differences. We conclude that intra-specific variability of otolith variables morfogeométricas *Prochilodus nigricans* showed that the shape of the otoliths were different between specimens from different geographical origins.

Abstract reference: IVE_Muntoni_24

Lapilli vs sagittae: results from the comparison of settlers of the striped red mullet

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Otolith microstructure resolved on a daily basis is a valuable tool to identify ecological and oceanographic processes influencing the early life of marine fishes and their recruitment. Counting otolith daily growth increments is time-consuming and, for some species, it lacks of accuracy and precision for estimating the early life larval traits. Most of the research has been carried out on sagittae or lapilli data, but the potential differences in the daily deposition between various otoliths' types have been poorly tested. To test differences on PLD estimations and to minimize preparation and counting errors associated with microstructural examination, we compared the microstructure of sagittae and lapilli of *Mullus surmuletus*, a commercial highly valuable and targeted species in the Mediterranean sea. The individual, ranging from 25 to 70 mm in total length, were caught during settlement, in coastal waters of the Western Mediterranean.

Abstract reference: IVE_Bellodi_25

Life history parameters of the small Mediterranean-endemic skate, *Raja polystigma* Regan 1923, from Sardinian seas

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The life history parameters of the speckled skate, *Raja polystigma* were investigated through the vertebral centra analysis. A total of 183 specimens (97 females, ranging from 11.5 to 59 cm in TL and 87 males with TL along 11.8 and 52.1 cm) were caught during experimental (MED.I.T.S.) and commercial trawl surveys in the period 2012-2013. The vertebrae centra extracted from a subsample of at least 10 individuals, belonging to different size classes, were employed to state which technique among the EDTA decalcification process, the analysis of unstained centra and two staining methods (Alizarin Red and Silver Nitrate), allowed a better band enhancement. The unstained centra and those treated with Alizarin red provided a better band visibility, so these techniques were applied to the entire sample. The annuli counts were carried out by two different readers and repeated two times for both treatments. The obtained readings showed a good reproducibility with both techniques, even though band counts executed on stained centra (PA = 86.8, %Cv = 7.0, IAPE = 5.177) seemed slightly more accurate than those made on unstained samples (PA = 86.6, %Cv = 7.9, IAPE = 5.96). These differences were statistically proved through the Kolmogorov-Smirnov test ($p < 0.005$). The von Bertalanffy Growth Functions were determined for

both stained and unstained samples (respectively $L_{inf} = 75.93$, s.e. = 7.29; $k = 0.10$, s.e. = 0.02; $t_0 = -2.29$, s.e. = 0.25; $L_{inf} = 74.97$, s.e. = 7.35; $k = 0.10$, s.e. = 0.02; $t_0 = -2.16$, s.e. = 0.24). Given these results is recommended to apply the Alizarin red staining methods for future age estimations, through vertebral centra, of the speckled skate, before trying other more expensive and less time-efficient techniques.

Abstract reference: IVE_Odelstrom_26

Life-history/age structure of three-spined sticklebacks in the Baltic Sea

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Three-spined stickleback is a species that has received considerable attention in behavioural ecology, while relatively little is known about its life cycle and role in the food web, especially in marine and estuarine systems. Available data suggests that sticklebacks have increased dramatically in abundance since the early 2000's in different parts of the Baltic Sea, and the species now constitutes a considerable part of the fish biomass of both offshore and coastal ecosystems. Sticklebacks have been shown to affect the dynamics of coastal ecosystems, by affecting biomasses of large predatory fish like pike and perch, as well as the production of ephemeral algae. Despite this, little is known on the life-history of sticklebacks in the Baltic Sea. In order to increase the knowledge base, we have made an attempt to age three-spined sticklebacks on the spawning grounds in coastal shallow bays and in offshore areas of the Baltic Sea.

A total of 155 individuals from the northern and central parts of the Baltic Sea were analyzed, and individual lengths were determined to the nearest millimetre. Grounded and polished otoliths were examined using a stereo microscope to determine the number of annual rings. Determining the position of the first annual ring was difficult due to "false" rings. To aid correct age determination, a method was developed where the first annual ring was identified by staining and counting of daily growth increments.

A high temporal variation in age and length was shown among sticklebacks caught at the coastal site, depending strongly on spawning season. In the offshore the length at age differed slightly between areas. Individuals from the northern part of the Baltic Sea were significantly larger, at the same age, compared to the fish from central part. The oldest individuals found in the northern area were 4 years and in the central area 3 years, some reaching a total length over 9 cm.

Abstract reference: IVE_Al-Anbouri_27

Otolith Based Age and Growth Studies on the Indian Oil Sardine, *Sardinella longiceps* Valenciennes, 1847 From Muscat, Sultanate of Oman.

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The age and growth of Indian oil sardine *Sardinella longiceps* in Muscat area was determined by

examining the sagittal otolith. The seasonal and sub-seasonal increments were observed and interpreted as daily and sub-daily increments respectively. The Von Bertalanffy growth model was well fitted to the length at age for both sexes pooled data while sub-daily rings excluded from analysis when reading the otolith increments. The growth parameters were estimated as $K = 5.0 \text{ yr}^{-1}$ and $L_{\text{inf}} = 250 \text{ mm (TL)}$. Similarly, the sub-daily rings were considered as daily rings in the analysis, representing a better fit to length age data and growth parameters which were obtained as $K = 2.4 \text{ yr}^{-1}$ and $L_{\text{inf}} = 235 \text{ mm (TL)}$. The study examined the two given analyses when counting without and with sub-daily increments. It showed some significant differences in age and growth rate of the given analyses between 7 and 16 months of life span of the species. The latter age was more appropriate and reasonable when counting total rings as this species seemed to live for more than a year as these species attained maturity at the end of one year.

Abstract reference: IVE_Chung_28

Otolith microchemistry reveals ontogenetic records of vertical migration and metabolism of four dominant deep-sea fish in the Northeast Atlantic

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Effective fishery management demands an appreciation of spatial and behavioral variations in fish life history but this is particularly challenging in inaccessible deep water species. The chronological properties of otoliths, coupled with stable isotope records of environmental and physiological conditions, offers an approach to reconstruct ontogenetic migrations and metabolic histories in deep-sea fishes. Here we derive ontogenetic records of stable oxygen and carbon isotopes from four contrasting species of deep-sea fishes (*Alepocephalus bairdii*, *Antimora rostrata*, *Coryphaenoides rupestris* and *Spectrunculus grandis*) and compare these with previous isotopic data in other species. Oxygen isotope records indicate that ontogenetic increases in depth are a common feature in deep water fishes, but the nature of these migrations varies between species. Larval shallow water phases are found in all investigated species other than the morid, *A. rostrata*. However, the duration of this shallow water phase varies from short (*Hoplostethus atlanticus*, *Cataetx laticeps* and *S. grandis*) to covering much of the juvenile life history (*C. rupestris* and *A. bairdii*). Some species demonstrate more complex but consistent horizontal (*Aphanopus carbo*) or vertical (*H. atlanticus*) migration patterns. An increase of otolith carbon isotope values in terms of a decrease of metabolism is observed from all species but in various magnitudes. This metabolic history is associated with temperature changes, fish growth rates and body mass, which factors have different influences on the metabolism according to fish vertical migration patterns and life history traits. Differences in ontogenetic behaviors indicated by otolith microchemistry have implications for the relative vulnerability of each species to anthropogenic disturbances.

Abstract reference: IVE_Murie_32

Spatial and Temporal Effects of the Deepwater Horizon Oil Spill on Growth of Spotted Seatrout and Red Drum in the Gulf of Mexico

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One of the most immediate fisheries concerns following the Deepwater Horizon (DWH) oil spill in the Gulf of Mexico in April 2010 was to what extent the productivity of recreational and commercial fisheries would be impacted. Our ongoing research focuses on this concern by comparing the growth of representative fish species before and after the DWH oil spill event, with additional comparison to a control area that was not physically impacted to the same extent (*i.e.*, following a Before-After-Control-Impact, or BACI, design). Spatial and temporal changes in the age-specific growth of fish prior to and after the DWH oil spill is being estimated using the growth increments in their otoliths, which record the entire growth history of the fish from birth to capture and span the 2010 oil spill event. Fish species chosen for this study are representative of different habitats (inshore estuarine areas, reefs, sand/mud, offshore waters) and trophic levels (detritivore, demersal carnivore, piscivore, pelagic carnivore), and include spotted seatrout, red drum, striped mullet, sheepshead, southern and Gulf flounder, red snapper, gag, gray snapper, greater amberjack, and king mackerel. We are currently focusing on spotted seatrout and red drum, as estuarine-dependent species. Any changes will then be incorporated into age-based stock production assessments to estimate changes in productivity. The impact of the DWH oil spill at a fisheries ecosystem level is being modeled using the historical growth of these fishes using otolith sclerochronology, where the widths of the growth increments (rings) are matched to known environmental variables. This will be used in combination with ARIMA models and intervention analysis to statistically evaluate impacts. This research will develop the framework to provide a better understanding of the relative impacts of environmental catastrophic events, such as the DWH oil spill, on growth and production of important coastal fish stocks in the Gulf of Mexico.

Abstract reference: IVE_Fotiadis_33

The thickness of daily rings statoliths of *Todaropsis eblanae* and the relationship between the month, age and sex

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The statoliths of cephalopods are always installed in pairs within two statocysts, which are at the back of the head, below the eyes and below the siphon. Examination of the microstructure can reveal a lot of information on the age and development of individuals. For this task they were removed from 79 individuals of the species *Todaropsis eblanae* (the specimens were collected from Ionian sea) their statoliths and once purified from tissues and brain fluids, processed so as to reveal the daily rings. After such processing we received images via camera connected with the computer and through the program image pro plus, counted the number of rings and the distances between

them. With these measurements was correlation between the thickness of the rings month by month, the age and sex of the individual. The results showed that all three of these parameters are statistically significant and affect the thickness of statoliths in differently ways.

Abstract reference: IVE_Albuquerque_34

Tracking the use of freshwater streams by juvenile mullets (*Mugil liza*) through the analysis of Sr:Ca ratios in otoliths

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Juvenile mullets (*Mugil liza*) are vastly found in both coastal freshwater streams (or washouts) and marine surf-zones along the southern Brazilian coast. Those streams interconnect the marine environment with freshwater wetlands located behind the foredune ridges. Our general goal is to evaluate the importance of the coastal streams to mullet's life cycle through the chemical analysis of their otoliths. The specific objective is to indentify chemical signatures in otoliths capable of distinguishing fish that used marine and coastal streams during their early life and evaluate core-to-edge chemical profiles as tools to understand habitat use and migration. We sampled 12 juveniles in a coastal stream (≈500 m far from the surf-zone) and 12 in the adjacent surf-zone, and analyzed their otoliths with LA-ICPMS for $^{86}\text{Sr}:$ ^{43}Ca , $^{138}\text{Ba}:$ ^{43}Ca , $^{24}\text{Mg}:$ ^{43}Ca and $^{55}\text{Mn}:$ ^{43}Ca ratios. Only Sr:Ca ratios presented consistent differences between habitats.

Abstract reference: IVE_Lill_35

Two-spot PIXE analyses of polished otoliths for identification of anadromous whitefish in the Baltic Sea

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European whitefish (*Coregonus lavaretus*) exists in two sympatric forms in the northern Baltic Sea; an anadromous river spawning and a sea spawning form. The groups form mixed populations outside breeding time and place. The river spawning whitefish in the northern Baltic Sea has diminished mainly due to anthropogenic destruction of the spawning rivers and the conditions in them, and is presently listed among endangered species on the HELCOM red list of Baltic Sea species in danger of becoming extinct (BSEP 140) and as a globally vulnerable species on the IUCN red list of threatened species. Methods for whitefish identification are a prerequisite for a sustainable management of the fish stocks. However, river spawning whitefish cannot be

distinguished from sea spawning whitefish based on outer morphology, and alternative methods are therefore searched for. In this work we describe an application of particle-induced X-ray emission (PIXE) to the elemental analysis of polished otoliths of whitefish. Two spot on polished otoliths were irradiated; one spot in the centre of the otolith and one in the periphery. The irradiations were performed in air with a collimated 0.5 mm proton beam. The ratio of the strontium concentrations in the two spots was used to distinguish between the different groups of whitefish. Criteria on the ratios were suggested for identification of the whitefish groups. The results were compared to results of μ -beam PIXE scans as well as multi-point scans with the 0.5 mm proton beam. The measuring set-up and the results are discussed.

Abstract reference: IVE_Prokhorova_37

Using of individual biological features as auxiliary indicators in the age reading

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It's very important to solve whether the hyaline ring on an otolith edge is a year ring of the previous year (individual growth has not begun yet) or of the current year (individual growth has finished). These doubts about age readings regarding to fish caught in summer can occur. The method of herring otolith age reading was detailed. Analyses of morphological and physiological features of winter ring formation and beginning of increment on otoliths of the Norwegian Spring-Spawning herring was carried. Ring formation begins first on the front edge and then on the back edge of an otolith. Finally rings form on the upper and lower edges of an otolith. Beginning of feeding is a factor which determines beginning of increment on herring otoliths. Significant fat deposition on internals causes stopping of feeding and, thus, it is a factor which beginning of winter ring formation. Quantity of fat deposited on internals and stomach filling can be used as an auxiliary indicator during age reading. When there are doubts about age reading (as to fish caught in summer) it is necessary to pay special attention to the state of investigated rings on the front and back edges of an otolith as well as it is necessary to take into account quantity of fat deposited on internals and stomach filling, as individuals, whose growth has finished in the current year, will have fat quantity of 2-3 balls and small stomach filling (0-1 balls). This helps to solve whether the growth of otoliths has finished.

Abstract reference: IVE_Pérez-Mayol_38

Were you a reared meagre? Marking of *Argyrosomus regius* otoliths to identify recaptures

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Fish restocking is a common practice used to introduce or re-establish fish populations in a certain environment, either for ecological or economical purposes. When target species for fisheries are restocked it is crucial to identify if landings appertain to reared or wild individuals; such a

differentiation can be accomplished by otolith marking of released individuals born in captivity. *Argyrosomus regius*, a valuable top predator, became very rare in the western Mediterranean, therefore a restocking programme has been established in Majorca Island. We present here several experiments to achieve bulk otolith-marking of laboratory-reared meagre using fluorescent dyes baths (OTC and ALZ) for juveniles and ^{137}Ba injection of progenitors. ^{137}Ba marking took into account the Ba capability to traverse the egg yolk-larvae barrier and be deposited in otolith primordium. Fluorescence in the juvenile otoliths was assessed by UV-light microscopy. Quantification of the light isotope Ba incorporation into otoliths was performed by the measurement of the ^{138}Ba : ^{137}Ba relationship using Solution-ICPMS. The feasibility of the marking techniques was established evaluating the survival, the marking success (% fish with clear mark signals) and changes in growth rate which could affect post-release survival. The cost of each marking technique was also assessed.

Slideshow Poster Session (IVSS)

Abstract reference: IVSS_Chang_01

Age and growth of *Chrysochir aureus* and *Otolithes ruber* in the southwestern water of Taiwan

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Chrysochir aureus and *Otolithes ruber* (Sciaenidae) distribute in the Indo-West Pacific and are of commercial importance for coastal fisheries. A total of 381 *C. aureus* and 136 *O. ruber* ranging 130-475 mm and 122-363 mm, respectively, were collected in the southwestern water of Taiwan between November 2012 and January 2014. The von Bertalanffy growth models of the fish were established by age determination of otolith. Marginal increment analysis indicated that the otolith annuli formed annually in winter for both species. Maximum observed age 5 yr with asymptotic length 525 mm, growth rate 0.33 yr⁻¹, and growth performance index 4.955 were estimated for *C. aureus*, whereas 3 yr, 334 mm, 0.42 yr⁻¹, 4.958 were for *O. ruber*. This indicated that *C. aureus* is of relatively slow growth but larger size than *O. ruber*.

Abstract reference: IVSS_Neil_02

Age validation of NZ Paua: the relationship between age and growth rings on the shell

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This project aimed to determine whether protein layers in the shells of paua (*Haliotis iris*) could reliably be used to estimate age in paua. The number of these rings in the shells of abalone has been used to estimate age in many overseas species; however, the literature is variable and conflicting. Oxygen isotope ratios were used to estimate the number of seasonal temperature cycles along the growing axis of the shell. The basis of this method is that variations in water temperature lead to a measurable change in the ¹⁸O/¹⁶O ratio of shell material (expressed as δ¹⁸O), and this relationship is believed to be approximately linear between about 5 and 30°C. These shells were then sectioned, and the number of protein layers within vertical sections of the shell were counted and compared with the number of temperature cycles estimated by oxygen isotope analyses. The shells examined came from three sites in Quota Management Area (QMA) PAU 5A and three sites in QMA PAU 3. There appeared to be a relationship between the number of protein layers in the shell and the age indicated by stable oxygen analyses in many of the shells examined; however, there was no evidence that the protein layers were laid down on an annual basis. A major problem is that the protein layers do not appear to be laid down in shells at the same time of year at the same

location. If these layers were always laid down at a similar time of year at sites, it may provide some evidence of seasonal or annual deposition. The New Zealand Ministry for Primary Industries Shellfish Stock Assessment Working Group has suggested that regular monitoring of a local population for a period extending beyond a year may provide evidence about the seasonality of deposition of the protein layer.

Abstract reference: IVSS_Loh_03

Otolith atlas of Malaysia

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Otolith morphology, a species-specific character for the identification of teleost fishes, is widely applied to various studies on taxonomy, systematics, fishery biology, trophic ecology, paleontology and archaeology. Although otolith atlases of various regions in the world's oceans have been published, there is still limited information on the otoliths of Malaysia fishes. The present study proposes to build up an otolith collection and otolith database for Malaysia fishes. Among the various uses of the otolith database, is its use to eventually produce an otolith atlas for the Malaysian fishes. This presentation illustrates and describes the morphological characters of sagittal otoliths obtained from more than 2000 local fishes belonging to 53 families and 122 species inhabiting the rivers, estuaries and coastal waters of Malaysia.

Abstract reference: IVSS_Zitek_04

Transgenerational marking of freshwater fish otoliths using enriched stable Sr isotopes

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Transgenerational isotopic marking has been recognized as an efficient tool for mass marking of high numbers of fish larvae by injecting female spawners with enriched isotope solutions. So far mainly enriched stable barium isotopes have been applied for this purpose. Here, we present an alternative approach for transgenerational marking using natural stable strontium (Sr) isotope spikes as non toxic alternative administered at natural Sr levels. The successful transfer of enriched stable Sr isotope single and double spikes (⁸⁴Sr and ⁸⁶Sr/⁸⁴Sr) from female spawners of brown trout (*Salmo trutta* f.f.) and common carp (*Cyprinus carpio* L.) to the centre of the otoliths of their offspring was studied by (laser ablation) multi collector inductively coupled plasma mass spectrometry ((LA)-MC ICP-MS). In the case of the ⁸⁶Sr/⁸⁴Sr double spike experiments, isotope pattern deconvolution (IPD) (applied to LA-MC ICP-MS data) allowed for the re-identification of

unambiguous individual-specific Sr isotope signatures in the otolith cores of the offspring. Enriched stable Sr isotope double spikes show great potential for transgenerational marking especially in freshwater systems, since absolute tracer doses needed to induce a significant mark in the otolith can be kept low and the use of a double spike allows for the variation of the composition resulting in a large number of individual distinguishable fingerprints. Details on the metrological protocol for data processing during isotope pattern deconvolution (IPD) are given in the presentation of Johanna Irrgeher *et al.*, Sr isotope pattern deconvolution of LA-MC ICP-MS data to detect individual-specific transgenerational marks in freshwater fish otoliths.

Abstract reference: IVSS_Piñeiro _05

How do otoliths grow? Linear daily growth increments and areas calculated in single-labeled otoliths from oxytetracycline-injected hake (*Merluccius merluccius*)

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Eleven otoliths of European hake *Merluccius merluccius*, tagged and injected with oxytetracycline at a known date and recaptured later, were studied. Otoliths were selected to provide a broad interval of post-injection time, ranging from 14 to 378 days. Oxytetracycline labels were detected from transverse sections examined with epifluorescence microscopy. Multiple fields were recorded and image montages from sections were used to contour the perimeter of the otolith and the label perimeter, the latter visualized as a fluorescent line. To estimate otolith daily growth rate, increments per day along the major axis were evaluated against the time since the label was administered. Area measurements of the otolith's shape at the time of labeling were made by virtually transforming back the shape based on the estimated growth rate. Interestingly, linear measurements were more variable than surface measurements. Taken together, these initial data suggest that otolith growth is better described geometrically using area measurements than linear measurements, which may provide new insights to age estimation in fish.

Age Validation Workshop

Time	Moderator	Presenter	Title
09:00	Richard McBride	Richard McBride	<i>Opening remarks</i>
09:05	WSAgeOral Kastelle_08	Craig Kastelle	Use of the stable oxygen isotope, ^{18}O , in otoliths as an indicator of fish life history events and age validation
09:25	WSAgeOral Andrews_13	Allen Andrews	Refined bomb radiocarbon dating of two iconic fishes of the Great Barrier Reef
09:55	WSAgeOral Hamel_10	Owen Hamel	Advances in Ageing Techniques and Age Interpretation for U.S. West Coast Groundfish
10:25			<i>General discussion</i>
10:40	Break		
	Ole Thomas Albert		
11:00	WSAgeOral Uriarte_06	Andres Uriarte	Validation of age determination from Otoliths for the Anchovy in the Bay of Biscay.
11:20	WSAgeOral Carbonara_15	Pierluigi Carbonara	Holistic approach on the age validation for the red mullet (<i>Mullus barbatus</i>) in the Southern Adriatic Sea (Central Mediterranean)
11:40	WSAgeOral Hehir_04	Imelda Hehir	A question of winter growth: Interpretation of Quarter 4 Celtic Sea whiting otoliths
12:00	WSAgeOral Lampri_09	Paraskevi Niki Lampri	Can otolith weight be used as a trustworthy and quick predictor of age in <i>Pagellus erythrinus</i> (Pisces, Sparidae)?
12:20	WSAgeOral McBride_03	Richard McBride	Use and nonuse of tests of symmetry in age validation studies
12:50			<i>General discussion</i>
13:00	Lunch		
	Allen Andrews		
14:30	WSAgeOral Albert_16	Ole Thomas Albert	Validation of annual zones in Greenland halibut otoliths from recaptures of chemically marked fish
14:50	WSAgeOral Bank_11	Crista Bank	Age validation of monkfish (<i>Lophius americanus</i>)
15:10	WSAgeOral Spear_02	Natalie Spear	Age and growth validation of the common thresher shark (<i>Alopias vulpinus</i>) in the northeastern Pacific Ocean
15:30	WSAgeOral Aldanondo_05	Naroa Aldanondo	Validation of the first annual increment deposition in the otoliths of European anchovy (<i>Engraulis encrasicolus</i> L.) in the Bay of Biscay
15:50	Break		

Time	Moderator	Presenter	Title
	Allen Andrews		
16:20	WSAgeOral Ferreira_07	Susana Ferreira	Validation of daily increments in the otoliths of Atlanto-Iberian sardine larvae (<i>Sardina pilchardus</i> Walbaum, 1792) reared under different diets.
16:40			Oral posters
17:30		Allen Andrews	<i>General discussion and concluding remarks</i>
17:45	End		

Age Validation Workshop Oral Presentations

Abstract reference: WSAgeOral_Kastelle_08

Use of the stable oxygen isotope, ^{18}O , in otoliths as an indicator of fish life history events and age validation

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The isotopic or elemental content of otoliths provides a view into the life history of fish. The stable oxygen isotope ($\delta^{18}\text{O}$) in seawater is thought to be in equilibrium with marine calcium carbonate (CaCO_3) structures such as otoliths. We applied the principle that $\delta^{18}\text{O}$ variability in marine CaCO_3 is inversely related to water temperature. This presentation is an overview of what can be learned by microsampling otoliths and measuring $\delta^{18}\text{O}$ by mass spectrometry. We analyzed $\delta^{18}\text{O}$ from three species of fish from three regions in the North Pacific: Pacific cod (*Gadus macrocephalus*) from the Eastern Bering Sea, saffron cod (*Eleginus gracilis*) from the Chukchi Sea, and small yellow croaker (*Larimichthys polyactis*) from the Yellow Sea. Up to 10 microsamples were extracted from any one year's otolith deposition, and up to 42 microsamples from a 5 year old otolith, representing the life history of the fish. We confirmed the relationship between water temperature and $\delta^{18}\text{O}$ in the otoliths ($r^2 = 0.74$) using otoliths with a known temperature history. In the larger body of our study, we saw evidence of seasonal temperature fluctuations, ontogenetic migrations, and possibly a tool to investigate temperature trends over time. In exploited populations of Pacific cod, the life-history $\delta^{18}\text{O}$ signal provided a method of developing a more accurate age reading criteria and an age validation. A comparison between Pacific cod and saffron cod $\delta^{18}\text{O}$ signals indicated different life history strategies in terms of temperature preference and possibly differences in habitat usage.

Abstract reference: WSAgeOral_Andrews_13

Refined bomb radiocarbon dating of two iconic fishes of the Great Barrier Reef

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Refinements to the methodology of bomb radiocarbon dating made it possible to validate age estimates of the humphead wrasse (*Cheilinus undulatus*) and bumphead parrotfish (*Bolbometopon muricatum*). Age for these species have been estimated from presumed annual growth zones in otoliths to near 30 to 40 years, respectively. The validity of these estimates was tested using bomb radiocarbon dating on the small and fragile otoliths of these species, and provided an opportunity to refine the method using advanced technologies. A regional ¹⁴C reference record from hermatypic coral cores from the Great Barrier Reef was assembled and ¹⁴C measurements from extracted otolith cores of adult otoliths was successful. Validated ages supported the accuracy of growth-zone derived ages using sectioned sagittal otoliths and refinements made in this study indicate bomb radiocarbon dating can be used on other species with small otoliths.

Abstract reference: WSAgeOral_Hamel_10

Advances in Ageing Techniques and Age Interpretation for U.S. West Coast Groundfish

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Because growth patterns of many west coast groundfishes limit the informational value of length data, fish ages are very important inputs to our age-structured stock assessments. At the Northwest Fisheries Science Center, we conduct a variety of ageing studies, and we review three recent avenues of research: age validation, alternative ageing methods, and understanding ageing error. Age validation: We developed the first bomb radiocarbon reference chronology for the California Current, using known-age petrale sole (*Eopsetta jordani*). Petrale sole spend a substantial portion of their first year of life in areas subject to variable upwelling. This variable environment illustrates the importance of using reference curves for age validation that are region- and species-specific, whenever possible. Alternative ageing methods: Traditional age-reading methods are time-consuming for long-lived species. Based on initial success with two groundfishes, we explore the use of otolith weights for rapid age determination of long-lived groundfishes. We also explore the consistency of these relationships over time and space. Ageing error: The Pacific hake (*Merluccius productus*) stock is characterized by infrequent, strong year-classes, surrounded by average and below-average cohorts. Ageing is conducted annually, such that readers routinely know the year of

collection. Ageing error is typically assumed to be largely consistent across years in stock assessments, however, we hypothesized that readers are more likely to assign uncertain hake reads to predominant ages. We conducted a double-blind study wherein previously read otoliths from many years were reread without readers knowing the collection year. Results confirmed that strong year classes experienced less effective ageing error in the regular course of ageing otoliths. Accounting for this tendency improved model fits to age data. Each of these research avenues has improved our understanding and is enabling us to develop more reliable population models and management guidance.

Abstract reference: WSAgeOral_Uriarte_06

Validation of age determination from Otoliths for the Anchovy in the Bay of Biscay.

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Comprehension of the annual pattern of annulus formation throughout the anchovy life span was first achieved from the observations of the strong 1982 year class which showed a neat annual progression of modal lengths passing through the fishery until the exceptional age of 5. Validation of the proposed method was subsequently obtained through monitoring of the progression of the strong 1987, 1989 and 1991 year-classes, both by spring annual surveys and by continuous sampling of the commercial catches, coupled to the monitoring of the seasonal marginal edge formation of the otoliths. Since then Age validation has been confirmed by the correlation between the pulses of recruitments (at age 1), as reflected in their relative occurrence in the population in Spring, and the abundance of those recruitments according to surveys.

Typically, annual growth of anchovy otoliths of the one and two years old diminish to about 2/3-1/2 and 1/3 of that occurring in their previous ages respectively. Growth of older ages (three and four) are rather similar as, or slightly lesser than, at age 2. Maximum growth (white band formation) occurs in summer and growth detentions (with translucent annulus formation) in winter time. However the opaque edge formation begins sooner at the age of 1 (around February-March) than at older ages (May or June). During the first winter several translucent rings are occasionally formed resulting in a composite annulus formation. In addition during June/July, at peak spawning, a check is formed in many of the one year old anchovies. However, not all year classes, neither all anchovies lay down the same amount of checks and many of them may not show any. As such age determination requires the knowledge of the typical annual growth pattern of otoliths, of their seasonal edge formation by ages and of the most typical checks.

Holistic approach on the age validation for the red mullet (*Mullus barbatus*) in the Southern Adriatic Sea (Central Mediterranean)

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The growth of *Mullus barbatus*, one of the most important fishery resource in the Mediterranean, has been studied using different methods, but few papers, among the many published in the last decade, were focused on age validation. This paper represents an attempt of ageing validation using different methods in an integrated approach. The uncertainty of ageing red mullet by otolith reading is mostly linked to the number of false rings laid down before the first winter-ring. In our trawl surveys samplings (MEDITS), the occurrence of red mullets in early life stage allowed to determine at 4 cm Total Length (TL) the size of the metamorphosis from the pelagic (blue) to the bottom (red) phase. The back-calculated size of the first ring corresponded to this length, then it was possible to define as “demersal” this ring. By both the marginal and increment monthly analyses on the adults (>10 cm TL) the deposition of a yearly annulus (June-November opaque, December-May transparent ring) was identified. The same monthly deposition pattern was observed in the recruits (<8 cm TL), corroborating the hypothesis of the deposition of one check (demersal) only before the first winter-ring. The Length-Frequency-Distributions (LFD) from the winter trawl survey were analysed and modal components (Bhattacharya method) identified. The averages of each mode were statistical compared with the results of the back-calculated winter-rings and the absence of significant ($p > 0.05$) differences confirmed that the two independent methodologies described the same age pattern. Moreover the von-Bertalanffy growth curves calculated by otolith reading and Modal-Progression-Analysis didn't show significant differences (Chen-test $p < 0.05$). Results from different methods combined with the early life stage observations, allowed to elucidate some controversial aspects of the red mullet otolith ageing, especially when the classic validation method (e.g. mark-recapture, captivity rearing, radiochemical dating) are difficult to implement.

Abstract reference: WSAgeOral_Hehir_04

A question of winter growth: Interpretation of Quarter 4 Celtic Sea whiting otoliths

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Whiting is regarded as one of the most difficult gadoid species to age, owing to difficulties in distinguishing true annual zones from other zones in the otolith structure. These problems, identified in 2005 (Whiting Workshop, CEFAS, UK), have not been resolved. Ongoing difficulties have been encountered in particular when estimating the age of Q4 otoliths. The difficulty with Q4 otoliths is deciding whether or not to count a translucent (winter growth) zone forming on the edge which may be either late or early in being laid down. This inconsistency makes it hard to distinguish whether a translucent zone at the edge is this year's or the previous year's growth thus making a year's difference in the ageing. Accurate ageing is important, as quarterly based Age-Length Keys are used to estimate the catch numbers at age for analytical stock assessment. In the past there are apparent inconsistencies in the cohort tracking of strong and weak year classes in the catch at age matrix. This can undermine the performance of the stock assessment models. This study investigates (1) the growth pattern in Q4 and Q1 and (2) any evident changes over time in the formation of the translucent zone. A mini international exchange using WebGR is co-ordinated by the Marine Institute, Ireland. This study encompasses the years 2004 - 2013 for Q4 whiting otoliths in ICES area VIIg to investigate the agreement on age readings for age groups 3 and 4, which make up the majority of the landings. In addition, an examination of the marginal translucent edge by age group will be investigated.

Abstract reference: WSAgeOral_Lampri_09

Can otolith weight be used as a trustworthy and quick predictor of age in *Pagellus erythrinus* (Pisces, Sparidae)?

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The aim of this study is to examine the nature of the relationship between otolith weight (OW) and age of *Pagellus erythrinus*. In total, 181 specimens from the Southern Aegean Sea were collected within DCF programme in 2013; their total length (TL) ranging from 63 to 448 mm was measured. From each specimen, both otoliths were extracted, cleaned, photographed, weighted to the nearest 0.0001g and stored. No statistical differences were found between left and right otolith, thus the left was used for the analyses. Ages were estimated by counting the annuli on the images of the left otolith with specimens ranging in age from 0+ to 14 years. The general linear models (GLMs) were used to describe the relations between otolith weights (OW) along with the factors age, sex, maturity and the continuous variables TL, fish weight and the otolith radius (R). The results showed that OW was found to be statistically significantly related only with age, TL and R. The ANOVA

analysis denoted a statistically significant difference between the mean OW of the age groups, despite the observed overlapping among them. The multiplicative model was the most adequate to fit the OW-R and OW-TL relations. A strong linear relationship between OW and age was observed and this model was used to re-estimate the age of the examined individuals based on the OW values. The three criteria: sensitivity, specificity and accuracy were used to validate the classification performance of the OW-age model. The results indicated that otolith weight can potentially be used to estimate fish age effortlessly and economically or at least, be a useful tool for the improvement of the otolith age readings. In our study, 6.49% of the otoliths readings were corrected according to the corresponding otolith weights.

Abstract reference: WSAgeOral_McBride_03

Use and nonuse of tests of symmetry in age validation studies

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Tests of symmetry are ideal for age validation, because they evaluate a null hypothesis that disagreeing paired ages are unbiased with respect to agreeing paired ages. These tests were introduced to fish aging research in two papers, in 1995 and 1998, and since 2005 one or more tests of symmetry are cited regularly. There are three formulations: McNemar's (1947) maximally pooled test, Evans and Hoenig's (1998) diagonally pooled test, and Bowker's (1948) unpooled test. McNemar's test is most sensitive to even small differences on one side of the diagonal, if there are many cells where these differences accumulate. Evans and Hoenig's test is most sensitive to an imbalance projecting along at least one of the off-center diagonals. Bowker's test is most sensitive to just a few differences, even a single pair of cells, once the difference is large enough. Using R software, I simulated paired age data with four types of bias, at three levels of precision, to evaluate these tests. McNemar's test was redundant with Evans and Hoenig's test, except when it performed poorly in simulations with under- and overaging at different age classes. Bowker's test was the least likely to falsely reject the null hypothesis (Type I error) but also the least likely to detect bias (Type II error) when even modest levels of variability were simulated. Evans and Hoenig's test had the best performance overall, although it was not immune to reasonable Type I error rate and modest rates of Type II error. Evans and Hoenig's test is recommended as the primary test, and secondarily, Bowker's test in certain conditions. Tests of symmetry are also well suited for quality assurance when training new readers or quality control of specific samples.

Abstract reference: WSAgeOral_Albert_16

Validation of annual zones in Greenland halibut otoliths from recaptures of chemically marked fish

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Greenland halibut is difficult to age, and there are no commonly agreed ageing protocol although a recent ICES workshop considered two recently developed methods as promising. Especially the mid-age (age 5-10) sequence of zones are considered highly uncertain and stock assessments are much in need of resolving the issue. During 2006-2008 several thousand juvenile Northeast Arctic Greenland halibut were tagged and injected with oxytetracycline (OTC), a compound that remains in the blood for 1-2 weeks and incorporates in calcified tissue produced during this period, making a visible time stamp in the otoliths just after release. The fishes were released in the nursery area north of Svalbard and recaptured in both the Barents Sea and in Faroese and Icelandic waters. More than 40 recaptures that had been at large for 1-6 years were returned with their otoliths in place. Otolith growth increments from the periods at large were analysed with respect to zonation pattern and increment width. Results were discussed in relation to somatic growth increment, sex and recapture area. Based on the results, a modified ageing protocol is suggested.

Abstract reference: WSAgeOral_Bank_11

Age validation of monkfish (*Lophius americanus*)

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Monkfish, *Lophius americanus*, are an important component of commercial fisheries in the Northeast United States, but annual catch limits are precautionary, because of uncertainty in the stock assessment, including uncertain age determination. Annual growth rings are presumed to follow a seasonal pattern and are counted on the vertebrae to estimate age. However other calcified structures, illicia and otoliths have also been used to estimate age for other *Lophius* species. To validate annual growth rings on all three structures, monkfish were injected with a chemical marker, oxytetracycline or fluorexon, and kept alive for a year in the laboratory subjecting them to a seasonal cycle of temperature and light. Monkfish were also injected and released into the wild as part of a data storage tag study. The injected chemical leaves a visible mark on the growth ring that is being formed at the time of injection. After a year in the laboratory, monkfish were euthanized and digital images of sectioned vertebrae, illicia, and otoliths were captured under

ultraviolet light to illuminate the mark, and reflected light, which obscures the mark but shows the growth rings. Images under ultraviolet and reflected light were merged to reveal the location of the chemical mark and to count subsequent annuli. Results from both the laboratory holding study and the mark-recapture experiment indicate annual rings are evident on the vertebrae and illicia, but they are not identifiable on the otoliths.

Abstract reference: WSAgeOral_Spear_02

Age and growth validation of the common thresher shark (*Alopias vulpinus*) in the northeastern Pacific Ocean

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The common thresher shark (*Alopias vulpinus*) supports important commercial and recreational fisheries in the northeastern Pacific Ocean. The species is presumed to be relatively long-lived with females reaching maturity at an estimated 5-7 years, although age validation studies have not been conducted for the northeast Pacific population. In order to guide sustainable management decisions, a comprehensive understanding of the status of the population including their longevity and maturity schedule, is needed. The purpose of this study was to use oxytetracycline (OTC) injected animals to validate the band pair deposition rate in vertebrae. OTC is absorbed by calcified structures of the vertebrae so that time-at-large can be correlated to the number of band pairs deposited for direct validation. OTC tagging occurred off southern California from 1998-2012. A total of 2,137 thresher sharks have been tagged with conventional tags and 1,445 of these animals were injected with OTC ranging in size from 56 to 230 cm fork length (FL). Vertebrae from 54 (27 male, 27 female) OTC-tagged and recaptured animals ranging from 63 to 145 cm FL have been returned and 26 of these samples are from individuals at liberty for over one year with a maximum time at liberty of 1,389 days (3.8 years). Vertebral band pair deposition rates are currently being determined to validate annual growth patterns of the thresher shark at multiple life stages using X-ray image analysis. Preliminary results suggest one band pair per year at all size ranges collected. Marginal Increment Analysis (MIA) is used to assess periodicity of band formation, and Centrum Edge Analysis (CEA) to determine seasonal variance in growth. This information will be used to provide accurate age and growth models for the common thresher shark in the northeast Pacific Ocean and provide information necessary for accurate stock assessments.

Abstract reference: WSAgeOral_Aldanondo_05

Validation of the first annual increment deposition in the otoliths of European anchovy (*Engraulis encrasicolus* L.) in the Bay of Biscay

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Otolith macrostructure analysis enables fish age determination, which is fundamental to an understanding of their population dynamics. It is assumed that the annual growth increment,

known as annulus, is laid down at regular interval following a seasonal cycle. The opaque band formation is related to fast growth period, whereas the translucent band or “winter ring” is generally associated with a period of slow growth rate. However, in order to obtain reliable age estimates it is necessary to validate annual increment deposition in otoliths. For European anchovy, early juveniles were captured in October 2012 in the southern Bay of Biscay and maintained in captivity in a spherical culture cage until April 2013. Fifty juveniles were sampled at around 27-day intervals and measured for standard length. Otoliths were removed from each sample and processed following the methodology described for juveniles. From October 2012 to January 2013, anchovy juvenile lengths increased slightly or remained stable at around 98 ± 6.3 mm (mean \pm S.D). After this period, standard length significantly increased up to the mean value of 120 mm in April 2013. Likewise, the age (in days) of anchovy juveniles was estimated based on otolith microstructure analysis. The estimated age varied from 96 (for individuals sampled in October 2013) to 293 days (for anchovies sampled in April 2013). The general otolith daily growth pattern showed that increment widths increased rapidly and were broadest between 62 and 68 days, with mean \pm S.D of 20.1 ± 6.2 μ m. Thereafter, the widths decreased steadily to 1.5 μ m. This study also revealed that, in the case of European anchovy in the Bay of Biscay, the first translucent band formation started in October-November and it was completed in April.

Abstract reference: WSAgeOral_Ferreira_07

Validation of daily increments in the otoliths of Atlanto-Iberian sardine larvae (*Sardina pilchardus* Walbaum, 1792) reared under different diets.

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Ageing technique assuming daily otolith deposition increments has been widely used for fish larvae, but studies validating this technique are relatively rare. In order to validate the daily increment deposition in otoliths microstructure, sardine (*Sardina pilchardus*), larvae were reared in laboratory under starvation and with 3 different diets that included microalgae rotifers and *Acartia grani* and whose range of food densities translate into feeding and growth rates extending from less than required for maintenance (<2 prey L⁻¹) to saturated feeding levels (>5 prey per L⁻¹). Otoliths of 653 larvae from 0 to 40 days-post-hatch (dph) were successfully extracted and analysed. Growth increments, first-check and otolith diameter were determined in relation to age and total length. At hatching, the sagittae of each larvae consisted of a lenticular core with 10,49 μ m (\pm 1,05 μ m SD) diameter and a first check was formed with a diameter of 11,85 (\pm 1,45 μ m SD). An overestimation of larval age was recorded for some of the younger larvae (<5 dph), probably because of the presence of embryonic increments. Increment formation was less than 1 per day for larvae <15 dph and no significant differences was observed between high, intermediate or low food concentration whereas starved larvae showed significantly lower increment count when compared to the other diets. The slope of increments count (IC) vs age was much steeper for larvae older than 15 dph reared with high and intermediate concentration diets, but increment deposition was significantly less than 1 per day (0.8+0.102 IC). In conclusion, food concentration had a significant effect on increment deposition and otolith diameter for sardine larvae. These reveal the importance of being cautious when aging young and/or slow growing larvae at these temperature when sardine spawning peaks and larvae are most abundant off the Atlanto-Iberian coast.

Age Validation Workshop Posters

Abstract reference: WSAgePHastie_12

Upwelling impacts on a California Current bomb radiocarbon reference chronology and petrale sole (*Eopsetta jordani*) age validation

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We present the first bomb radiocarbon reference chronology for an eastern boundary current ecosystem, the California Current (CC), using known-age petrale sole (*Eopsetta jordani*) samples. We also present a validation study for petrale sole using this reference chronology. As petrale sole is a valuable groundfish harvested in the CC, proper ageing is important for its assessment and management. The validation study shows that petrale sole break-and-burn and surface ages are negatively biased by approximately 1 year and 2–3 years, respectively. However, after approximately age 10, the negative bias in surface ages increases rapidly and surface-read age estimates beyond approximately age 14 are entirely unreliable. Both the reference and validation curves are more variable and show a lag in the rate of radiocarbon increase, relative to most other time series of bomb radiocarbon in marine systems. Petrale sole spend a substantial portion of their first year of life in areas subject to variable upwelling, which drives the introduction and mixing of low-radiocarbon deep waters with surface waters that are more closely linked to atmospheric conditions. This study suggests that the variability and lagged rate of radiocarbon increase is most likely due to coastal upwelling, illustrating the importance of using reference curves for age validation that are region- and species-specific, whenever possible.

Abstract reference: WSAgeP_Barcala_07

Corroboration of faster annual growth rate of black anglerfish (*Lophius budegassa*) in Spanish Mediterranean based on length frequency analysis.

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The illicium (first dorsal fin ray) has become the standard structure for age estimation of black anglerfish (*Lophius budegassa*) in most of the European countries and the one used as a basis for stock assessment, after calibration exchanges among the age readers and workshops. However, the tendency in the past to not pay essential attention to the need to validate or corroborate the age estimation in some stocks has led to misunderstanding of their dynamics. Thus, the Atlantic black anglerfish stocks have not been annually assessed using age-structured models during the last years in ICES working groups due to problems with the data quality, including those coming from a biased illicia age estimation criterion. The same age estimation criterion is used for the Mediterranean black anglerfish. Evidence obtained in the last years, using alternative methodologies to the age

estimation, have shown a faster growth rate of black anglerfish. Some growth increments considered as annuli in illicia do not seem to be really annual.

The growth pattern of black anglerfish in Spanish Mediterranean waters is analyzed in this study using the length-frequencies (lengths ranging 3-128 cm) of annual groundfish research surveys between 2004 and 2012. The results corroborate that the growth pattern of the Mediterranean population of this species is also faster than that estimated applying the traditional illicia age estimation criterion. The von Bertalanffy growth parameters are also estimated and compared to those of previous studies; they will be available if a length-structured model including growth parameters is used in the assessment of this Mediterranean stock.

Abstract reference: WSAgeP_Baudouin_17

Comparison of otolith and scale readings for age estimation of common dentex (*Dentex dentex*)

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The common dentex, *Dentex dentex* (Linnaeus, 1758), is a demersal sparid fish (0-200 m), that grows to a maximum length of 100 cm and a weight of 13 kg, with a relatively long life span. It is classified as "Vulnerable" and considered to be threatened in the Mediterranean regional IUCN Red List. It is mainly targeted by both small-scale artisanal fisheries and recreational fishing, but there is currently very poor methodological and quantitative information about its age estimation despite its conservation status. This work aims to estimate the age and growth of common dentex in Corsica using two different calcified structures: the scales, which are the most frequently used for ageing because easily collected, and the otoliths, which are more difficult to collect but are known to be more reliable. Furthermore two methods were applied for otoliths: one using whole otoliths read in glycerol, and the other one using embedding and cross-section followed by Toluidine blue coloration. Thus, we compared three methods of age estimation for this long-lived species. The estimations were made by two independent readings from a principal reader and by a third reading from a secondary reader. Age determination from scales appears to be relevant only for young individuals up to four years, beyond this age the differences between readings were significant. Moreover the maximum estimated age with scales is 16 years. Alternatively, from whole otoliths, the age estimation is reliable up to 12 years, and the maximum estimated age is 27 years for whole otoliths and 30 years for sectioned ones. Validated ages through these three methods allowed determining the Von Bertalanffy growth parameters and characterizing the demographic structure of exploited individuals in Corsica. These results provide new information for age estimation in this species that will contribute to a better assessment of its population dynamics.

Abstract reference: WSAgeP_Bekas_28

Daily rings observations in blackspot seabream *Pagellus bogaraveo* (Brünnich, 1768)

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Daily rings have been used to estimate age and growth in blackspot seabream (*Pagellus bogaraveo*) juveniles. A sample of 46 specimens with total length (TL) ranging from 83 to 157 mm were collected from the Aegean Sea. From each specimen, both otoliths were extracted, cleaned, photographed and stored. The right otolith, was embedded in resin, sectioned, ground and polished. Slides of the sectioned otoliths were photographed and daily rings were counted using Image analysis software. The otolith nucleus indicated a central primordium. Estimated ages ranged between 172 up to 360 days. The first annulus seemed to be formed mainly between January and February. Daily rings were used for the ageing of the 0+ and 1+ age groups and the relationship between age in days and TL. These results are compared with the existing published works on *P. bogaraveo* ageing.

Abstract reference: WSAgeP_Blass_03

Comparing two methods of age determination of herring (*Clupea harengus*) in the Bothnian Sea and the Bothnian Bay

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In general, age determination of Bothnian sea herring is based on microscopic inspection of sectioned otolith. This method is quite expensive but considered to give reliable results. An alternate and cheaper method was tested using grinded transversal and stained otoliths. A test was set up in order to compare the two methods in terms of potential bias. The hypothesis was that the alternative method will underestimate number of growth zones of northern herring compared to the traditional method.

For the study a total of 90 herrings consisting of four samples from the the Bothnian Sea and two from the Bothnian Bay were prepared and read. The herring was selected from samples by hydroacoustic research surveys and commercial fisheries collected in spring and autumn 2013. From each individual one otolith (sagitta) was sectioned by a diamond saw and stained with neutral red. The other otolith was mounted on a polycarbonate chip with the centre of the nucleus at the edge of the chip and then grinded, polished and stained. This enabled a pairwise comparison of age determination methods.

Results showed that all readers estimated higher age from sectioned otoliths than from grinded. The difference was 1-2 years in mean age for the whole sample. For sectioned otoliths, variance of

the age estimate was higher than for grinded samples. Variances did not, however differ significantly. For both methods, variance increased with age (median for both methods). Between pairs of readers, the mean difference between age estimates was between 0,2-0,9 years for sectioned otoliths and between 0,1-1,1 for grinded otoliths. Age estimates from sectioned otoliths did not differ significantly for any pair of readers. Estimates from ground otoliths differed significantly for three pairs of readers. Sectioned otoliths seem more reliable, and microscope magnification may explain much of the difference.

Abstract reference: WSAgeP_Cerna_20

Age validation of the jack mackerel (*Trachurus murphyi*) off Chile

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The jack mackerel, *Trachurus murphy* Nichols 1920, is a very important commercial carangid in the southern Pacific Ocean. Despite their importance age determination is still a controversial issue in this species. To date jack mackerel ageing, achieved by reading annual rings in whole and sectioned otoliths, has been used for stock assessment from 1970s. The current indirect age validation of jack mackerel through the frequency of formation of growth increments using the edge analysis method seems to be necessary but insufficient to obtain an accurate age determination for this species. In the present study the first annulus in juveniles *T. murphy* was validated using daily micro-increment of sagittal otoliths. The accuracy of ageing was also validated through bomb-radiocarbon assay of 13 otolith cores whose year of formation ranged from 1959 to 1973. We compared the jack mackerel radiocarbon assay with the Pacific halibut scaled to jack mackerel reference chronology. This indicated that some of the jack mackerel section and whole-otolith ages were correct at least on average. The approximate mean age estimated from range of birth year based on delta ¹⁴C chronology was projected over von Bertalanffy growth curve. The results showed that the radiocarbon age of 13 fish matched to growth curve estimated from whole otoliths and confirmed that age from 40-55 cm FL jack mackerel ranged from 10 to 15 years. The age validation in jack mackerel will provide a more accurate estimation of growth parameters of von Bertalanffy and age-length keys for stock assessment purposed.

Abstract reference: WSAgeP_Chatzisyrou_24

Using otolith weight to improve age estimation in red mullet *Mullus barbatus*

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Otolith weight has been examined last decades as an alternative method for the ageing of fishes.

The current work investigates the relationship between otolith weight and several fish variables with emphasis to the age in *Mullus barbatus*. A total of 342 specimens from the Aegean and Ionian Sea were collected and measured ranging in total length (TL) from 94 to 229 mm. Otoliths were extracted, cleaned, photographed, weighted to the nearest 0.0001 g and stored. Broken or unreadable otoliths were excluded from the analyses. Since no statistical significant difference was found between the weight of the left and right otolith, the left one was used for the analyses. Ageing was based on the identification of annuli on digitized pictures by using Image Analysis Pro plus System, taking as date of birth the first of June. Ages ranged between 0+ and 6 years. The general linear models were used to describe the relations between otolith weight (OW) along with the factors age, area, sex, maturity and the continuous variables TL, fish weight and otolith radius (R). The results showed that OW was statistically significantly related with age, area, TL and R. As a result the analyses were performed for each area separately. Linear model best described the OW-R and OW-TL relationships in the Aegean, whereas the multiplicative model explained better these relations in the Ionian. OW was linearly related with age for both areas. OW-age model was used to re-estimate the age of the examined individuals based on their OW. Sensitivity, specificity and accuracy criteria were used to validate the classification performance of the OW-age model. ANOVA analysis indicated significantly different mean OW for each age group despite the overlapping among them. Overall, our results suggested that OW can be used to improve objectivity and accuracy in ageing of *Mullus barbatus*.

Abstract reference: WSAgeP_Freshwater_15

Validation of daily increments and a marine entry check in juvenile Sockeye Salmon using experimental rearing and microchemical techniques

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Accurate estimates of age and growth from otoliths depend on a strong correlation between otolith and somatic growth, the deposition of increments at a known rate, and the universal formation of marks of interest in sampled populations. Previous studies have indicated these assumptions do not hold for all species or age classes. We evaluated the formation of daily increments and a marine entry check in otoliths of 110 lab-reared, juvenile Sockeye Salmon (*Oncorhynchus nerka*) over a 100-day period. By smolting individuals in controlled conditions, we successfully created a distinct marine entry check that was used to enumerate the number of micro-increments after a known date. We also used inductively coupled plasma mass spectrometry (ICP-MS) analysis to assess visual bias and secondarily validate the formation of marine entry checks in a subset of wild-caught juvenile Sockeye Salmon. Otolith width and body size were strongly correlated in experimentally reared juveniles, while counts of micro-increments confirmed that they were deposited daily. In wild-caught juveniles, strontium concentrations could be used to effectively estimate marine entry. However, chemical estimates were often significantly earlier than visual estimates, particularly in individuals with gradual changes in elemental concentrations. The uncoupling between visual and chemical estimates suggests that changes to otolith microstructure may occur at different rates than the incorporation of metal ions. Pairing microstructure and microchemical techniques with detailed knowledge of water quality at study sites may provide increased accuracy when evaluating diadromous migrations.

Abstract reference: WSAgeP_Gregg_01

Clarity and accuracy of ageing transverse sectioned otoliths from black sea bass, *Centropristis striata*, from the Northwest Atlantic Ocean

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Accuracy in ageing is imperative for understanding population dynamics and for assessing the status of a stock. Uncertainties associated with recent assessments for the Northern stock of black sea bass, *Centropristis striata*, a commercially and recreationally valued species, warrant a reexamination of current ageing methods. For this study, scales and sagittal otoliths were collected from sites sampled in Southern New England and the Mid-Atlantic Bight by the Northeast Area Monitoring and Assessment Program (NEAMAP). Calcified structures, which typically display seasonal growth patterns interpreted as annuli, were analyzed without information regarding capture location, time, and fish size to minimize bias in age assignments. These preliminary studies compared various preparation methods and orientations of both otoliths and scales under a standard set of criteria. Structures were evaluated on mark clarity, reader agreement, structure growth, and processing efficiency. Future work will include year-round sampling for validation of the preferred structure. These studies will yield samples with enhanced clarity, resulting in more accurate age data for black sea bass and improved estimates of age-specific parameters considered for inclusion in the stock assessments of this species.

Abstract reference: WSAgeP_Hernández_11

Estimation of age and growth of juveniles of two European anglerfishes, *Lophius budegassa* and *L. piscatorius*, in the north-eastern Atlantic waters, from otolith microstructure analysis.

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The two European anglerfish, *Lophius budegassa* and *L. piscatorius* are commercially important species in Europe, but our understanding of their basic biology is far from complete, hence the need to investigate its early life history through the analysis of otolith microstructure. Otolith microstructure analysis is an useful tool that provides information for determining important early life history associated with age and growth of juvenile fish, such as hatch dates and settlement dates, and reproductive and recruitment patterns. The otolith microstructure can also provide valuable information on the frequency of the first annual increments formation that can be corroborated by counts of daily increments. Age, growth and hatching patterns of both European anglerfish species are here estimated by using lapilli microstructure analysis. The otolith microstructure of juvenile anglerfish (< 30 cm) captured from bottom trawl surveys in southern Bay of Biscay (ICES Div. VIIIc, IXa) in autumn is fully described. Analysis of the otoliths provided estimates of post-settlement ages, sizes at settlement and juvenile growth rates, enabling the back-calculation of the hatch date. This study represents the first attempt at characterizing the early life history of both European anglerfish, contributing also to corroborate the time and position of its

first annulus and thus to help set a more accurate annual age estimation criterion. This is essential for the stock assessment process of these species by using age-structured models, given the problematic of its annual age estimation.

Abstract reference: WSAgeP_Johansson_19

Multicriteria approach for validating the first winter ring deposition in Eastern North Sea plaice (*Pleuronectes platessa*) otolith: preliminary study

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A major difficulty in accurate age determination of plaice (*Pleuronectes platessa*) consists in how to interpret the first hyaline ring, sometimes read as a "settling ring" and sometimes read as a true annual ring. As the misinterpretation of this first ring leads to a bias in age based stock assessment, validating this first ring is crucial. The sampling was carried out between May and December 2011 in shallow water (2-5 m) in the Swedish fjord "Gullmaren", known to be a good settling ground. Here we applied a multicriteria approach based on edge analysis and morphometric measurements, useful in ring deposition validation studies when classic methods (e.g. mark and recapture, captivity rearing, radiochemical dating, bomb radiocarbon etc.) are difficult to implement. The marginal analysis of the otolith from adults (age>0) and juveniles (age 0) show the same pattern with a prevalence of the opaque edge in summer/ early autumn and transparent (hyaline) edge in late autumn. These results suggest that a false ring deposition before the first winter ring does not occur in juveniles. Moreover morphometric measurements of the radius in otolith with hyaline edge observed in specimens caught in October / November did not show significant differences (Wilcoxon-Mann-Whitney test; p>0.05) from the first ring displayed by adult specimens. Taken together both approaches present preliminary evidence indicating that the first ring in this plaice stock otoliths is likely to be the first winter ring.

Abstract reference: WSAgeP_Kashava_25

Preparation of hake *Merluccius* spp. otoliths for routine annual age determination in Namibia

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The two species of hake are the most important commercial resource in the northern Benguela upwelling system. Their ages are routinely determined on otoliths using the whole otolith method. The age data are used one of the main input datasets into the routine stock assessment used as the main consideration on which fisheries management strategies are built. The two hake species, the shallow-water hake *Merluccius capensis* and the deepwater hake *M. paradoxus* have thin enough otoliths to be able to accurately read the edges for annual age determination. However, on otoliths

> 30 cm fish, the centre becomes thick and some nuclear zones become difficult to read. In this paper, we address this issue by describing and testing the method of polishing. The aim of the method to thin down the otolith, especially on the proximal and rostrum surfaces, and we propose here, is similar to in accuracy for routine age determination purposes, and more cost-effective than the thin slicing method. Both otoliths of 50 fish > 30 cm were extracted. One otolith per fish was polished on a grit paper attached to a turntable of a polishing machine and water is pored over the grit paper to make it wet. The otolith is first polished holding down the rostrum (which is thicker) and polishing for approximately one minute and then polishing on the post rostrum for another minute. Otoliths are stored in vials of water for two days prior to polishing. Both the polished and the unpolished otoliths were read at different times and measured and numbers compared. Results show that polishing enhances viewing of the growth zones. In 20% of the otoliths the polished version reflected more translucent zones than the unpolished version.

Abstract reference: WSAgeP_Kousteni_26

Potential use of otolith weight to estimate stripped mullet *Mullus surmuletus* age

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The possibility of using otolith weight in the stripped mullet *Mullus surmuletus* ageing was examined. In total, 228 specimens from 2 areas were collected within DCF Program in 2013; their total length (TL) was measured to the nearest 1 mm. From each specimen, both otoliths were extracted, cleaned, weighted to the nearest 0.0001 g, photographed and stored. Broken or unreadable otoliths were excluded from the analyses. Ageing was based on the identification of annuli on the images of the left otolith using Image analysis software and taking as date of birth the first of June. Six age groups were identified (from 0+ to 5+). No statistical differences were found between the weight of the left and right otolith, thus the left otolith was used for the analyses. The relation of otolith weight (OW) with the variables age, area, sex, maturity, TL, fish weight and otolith radius (R) was examined using general linear models. OW was found to be statistically significantly related with age, TL and R. The relationships of OW-R and OW-TL followed the multiplicative model; however, OW was linearly related with age. The model of OW-age was used to re-estimate the age of the examined individuals from their OWs. Sensitivity, specificity and accuracy were examined to validate the classification performance of the OW-age model, showing promising results for the otolith weight based ageing of the stripped mullet. Furthermore, ANOVA analysis indicated significantly different mean OW among the age groups, although overlapping was obvious. The use of otolith weight is also proposed for the improvement of the otolith reading based ageing.

Abstract reference: WSAgeP_Lampri_14

Daily and annual rings for age validation in common pandora *Pagellus erythrinus* (Linnaeus, 1758)

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The possibility, that otolith microstructure can provide an accurate estimate of age and growth in common pandora (*Pagellus erythrinus*), was investigated in this study. A sample of 50 specimens with total length (TL) ranging from 63 to 160 mm were collected from the Southern Aegean Sea within DCF programme in 2013-2014. From each specimen, both otoliths were extracted, cleaned, weighted to the nearest 0.0001 g, photographed and stored. The annual rings were counted based on the images taken from the right otoliths using Image analysis Pro Plus software. Afterwards, the right otoliths were embedded in resin, sectioned and polished. Images of the sectioned otoliths were used for the daily rings identification. The otolith nucleus of most specimens displayed a common pattern with a distinct central primordium. Daily rings were used for validation of the 0+ and 1+ age groups and the relationship between age in days and TL was examined. Estimated ages ranged between 98 up to 726 days. The first annulus starts to appear at the end of April and seems to be fully formed by the end of May. The smallest individual which was observed to have fully formed the annual ring was 114 mm (TL). Estimated ages based on daily readings revealed a good agreement with the estimated ages (0+ - 2+) based on the identification of annuli counts macroscopically. These results can broaden the existed knowledge on growth and early-life ecology of this species and can be used for age validation.

Abstract reference: WSAgeP_Landa_02

Age estimation of megrim (*Lepidorhombus whiffiagonis*) corroborated by cohort tracking in northern Iberian waters

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To confirm the consistency of the age interpretation by validation studies is essential for providing accurate age estimates to the stock assessment process using age-structured models. The Atlantic Iberian megrim (*Lepidorhombus whiffiagonis*) stock is annually assessed by age-structured models in ICES, and specimens are routinely aged by IEO expert readers for this purpose. A total of 8845 otoliths of this stock were aged from 23 annual groundfish surveys carried out in north and northwestern Iberian waters (ICES Div. VIIIc, IXa) and following internationally standardized protocols. Yearly age-length-keys were built using the respective age estimates and applied to the length distribution of each survey, thus obtaining a matrix of abundance indices by age and year. Similar mean lengths are obtained for each age group along the time series. The annual age estimation of *L. whiffiagonis* is here indirectly validated or corroborated by tracking cohort abundance indices in the surveys. Strong and weak cohorts are well tracked along the most

of their age classes, and the correlations between age abundance indices are statistically significant up to the age 5, which represent around 94% of the total number of this species caught in the surveys. The von Bertalanffy growth parameters for the time-series are estimated (L_{∞} : 56; k : 0.12; t_0 : -1.88) and the results are compared with previous studies.

Abstract reference: WSAgeP_Lourenço_27

Precision in age estimates in *Octopus vulgaris* stylet increments analysis

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Octopus vulgaris (Cephalopoda) is a fast growing species with short life span estimated between one and two years. According with studies conducted in captivity and with animals captured in the wild, the species grows according with a exponential or power model with high individual variability. With recent improvements in preparation methodologies and the validation of daily increments deposition in the stylets (vestigial shell) of merobenthic and holobenthic octopus, the stylet increment analysis (SIA) achieved a step forward for the accurate age estimates in commercial and non-commercial species of octopus species. With the increasing use of SIA to estimate the age of wild populations of octopus worldwide, concerns with age estimates precision, repeatability of counts and comparison between studies arise. Herein, we analysed the precision of age estimations in stylets of *O. vulgaris* juvenile of the Portuguese Northwest coast based on the validation of the nucleus diameter conducted in newly-hatched paralarvae. Accordingly with our estimates juvenile octopus seem to take around 8 months to achieve the legal landing weight (750 gr). The precision levels obtained and the doubts raised in relation to the accuracy in individual age reveal the necessity to revalidate the deposition of daily increments in *Octopus vulgaris* particularly in pre and post settled juvenile.

Abstract reference: WSAgeP_Megalofonou_22

Age and growth of Atlantic bluefin tuna using otolith microstructure and macrostructure: The case study of the Mediterranean fish

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One of the main problems in the attempt to estimate age and growth is the selection of the most suitable structure to age the fish. Comparative analyses of different calcified structures and techniques in bluefin tuna suggested that spine and otolith sections are the most reliable. However, it is still uncertain whether the remarkable differences on mean lengths at ages observed among the studies may be attributed to the diversity of ageing methodology used, the geographic origin of the samples or both. This is the first attempt to estimate age and growth of Atlantic bluefin tuna

from the Mediterranean Sea using otoliths. Our main goal was to evaluate the precision of the method and compare the age estimates with results from previous independent studies that were based on either otoliths or dorsal fin spine sections. Sagittal otoliths were obtained from 278 bluefin tuna caught in the Mediterranean Sea during the period 1993-2008. In particular, a part of otoliths were obtained from specimens collected from the catches of professional fishing vessels, at landing or after tuna were transported to the wholesale market, while an additional sample was obtained from reared in sea cages specimens at Bluefin Tuna Farms. Specimens' size ranged from 8.5 to 278 cm in total length and from 7.5 g to 386 kg in total weight. Otoliths from juvenile specimens, smaller than 60 cm in fork length, were aged by enumeration of daily growth increments while otoliths from specimens larger than 60 cm were aged by enumeration of annual growth bands in otolith sections. The age estimates ranged from 20 days to 21 years and the von Bertalanffy growth parameters were determined ($L_{\infty}=330,8$ cm, $k=0,084$ and $t_0=-1,452$ years). The potential longevity of the species was found to be 35.7 years.

Abstract reference: WSAgeP_Montanini_08

Validation of first annulus formation in Triglidae family (Teleostei, Scorpaeniformes): a tool for increase the biological knowledge of a demersal resource

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Gurnards are demersal fishes exploited by commercial trawl fisheries that spend a pelagic phase during their early life history stages. There are eight different species in the Mediterranean showing intra- and inter-specific differences related to feeding habits, depth distribution and biological cycle. Although these changes have been widely reported little is known about their first life stages due to a lack of information on age and size at settlement to the bottom, length at first year of age and otolith formation. The present study is the first attempt to determine the first growth stages of six gurnard species: *Chelidonichthys cuculus*, *C. lucerna*, *Eutrigla gurnardus*, *Lepidotrigla cavillone*, *L. dieuzeidei* and *Trigloporus lastoviza* and to determine the first annuli formation. Indirect validation is made by counting presumed daily increments on two defined radii: the first radius is measured along the proximal-distal axis from the primordium to the last increment before the apparition of an accessory growth centre (AGC); the second radius is measured from the more dorsal AGC to the proximal edge of the otolith close to the sulcus acusticus and perpendicular to the increment deposition. The duration of the pelagic life phase was determined and the first annulus was defined by the position of one year-increment count in terms of distance from primordium and annulus density (*i.e.*, translucent vs. opaque). The location of discontinuous and incremental zones linked to developmental changes, the description of the species-specific patterns, as well as the measurement of increment widths are discussed in the light of the species ecology and biology.

Abstract reference: WSAgeP_Muñoz Rubio_16

Application of quality control ageing of *Trachurus murphyi* and *Merluccius gayi gayi* through procedures approved by ISO 17025

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The application of standardized protocols in age determination has been an important change to ensure quality control studies and also allowing documented procedures to facilitate continuous improvement. In the Age and Growth Laboratory of the Instituto de Fomento Pesquero (IFOP) a Quality Control Manual was developed under ISO 17025 whose primary objective is the accreditation of competition in the samples studied for two fishery resources, *Trachurus murphyi* and *Merluccius gayi gayi*. The Quality Control Manual consists of administrative and technical procedures, indicating the path to be followed in systematically developed trials, containing fifteen specific procedures, seven instructional and thirty-one forms. This documentation ensures the mechanisms and procedures of quality control in sample taking, age determination, including the validation of the accuracy and precision of the method of age determination, procedures for staff training and new analysts, use of reference collections and reading protocols of age, record checks on equipment and staff knowledge in respect of the requirements of the standards. From a technical point of view, the main documents are the "Quality Assurance Process" of age determination (Specific Procedure 8-4), which includes exercises of measurement for reproducibility, repeatability and the frequency with which these exercises should be carried out according to the experience of the analyst and the document containing the "Method of Age Determination" Specific Procedure (PE-8-5) that outlines the steps to be followed by the analyst. All documentation is in an intranet system, allowing easy access and allowing online updates without handling printed documentation which is obsolete. For its functioning, this system requires a high level of commitment from those doing the technical work, as well as the strata managers of the institution, (IFOP www.ifop.cl) groups who direct the management and administration of resources, this being a constant challenge to maintaining functionality of the quality control system.

Abstract reference: WSAgeP_Mytilineou_18

Assessing the otolith weight based ageing in picarel *Spicara smaris* (Osteichthyes: Centracanthidae)

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The present work evaluates the usefulness of otolith weight in *Spicara smaris* ageing. In total, 215 specimens from 2 areas were collected in the framework of DCF Program 2013 and their total length (TL) was measured to the mm. From each specimen, both otoliths were extracted, cleaned,

photographed, weighted to the nearest 0.0001 g and stored. Ageing was based on the identification of annuli on the images of the left otolith using Image analysis and taking as date of birthday the first of May. No statistical differences were found between the left and right otolith weight, thus the left otolith was used for the analyses. Otolith weight (OW) was examined to identify the relationship with factors such as area, sex and maturity and the continuous variables age, TL and weight of fish and otolith radius (R) using General Linear Models. OW was found to be significantly related ($p < 0.05$) only with fish age, TL and weight and otolith R. The relationships of OW-R and OW-TL fitted the model $Y = aX^b$. ANOVA analysis showed that mean OW by age group differed significantly among them. OW was linearly related with age. Re-estimation of the age of the examined individuals was performed based on the OW-age model and the individual OWs. The hypothesis that OW can be used to estimate the age was based on the criteria of sensitivity, specificity and accuracy for the evaluation of the classification performance of the model OW-age. The results showed that OW can be used for *Spicara smaris* ageing. Furthermore, OW was used for the improvement of the annuli readings from the otoliths. A relatively low percentage (2.3%) of the age readings were corrected after re-reading of the otoliths corresponding to unusual points in the ANOVA analysis and/or the OW-age model.

Abstract reference: WSAgeP_Navarro_10

First attempt to validate the age estimation of chub mackerel (*Scomber colias*) in the Bay of Biscay using otoliths

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The Atlantic chub mackerel (*Scomber colias*) is a pelagic fish distributed in warm and temperate Atlantic waters and in the Mediterranean Sea that may be considered the southern congener of Atlantic mackerel (*S. scombrus*), both species overlapping in Iberian waters.

Age determination is an essential feature in fish stock assessment to estimate the rates of mortalities and growth. Chub mackerel is not assessed, but its fishery has increased in northern Iberian Atlantic waters (ICES Div. VIIIc and IXa) and fishery advice of this population may be soon required. The increase in the landings may also reflect a shift in distribution further north possibly associated with climatic variation, and could interact with the population dynamics of other species. Therefore, it is important to know, as accurate as possible, the growth pattern of this species.

The age estimation criteria of chub mackerel are not still internationally standardized and have never been validated or corroborated in Iberian waters. This work presents the growth pattern in southern Bay of Biscay (ICES Div. VIIIc) based on samples from a whole annual period (2011). An attempt of semi-direct validation of the age estimation of this species in the area is performed, at first time, based on the otolith marginal increment and edge analyses. The consistency of the age interpretation is tested by the regularity of the increments formation and the comparison of the lengths at age obtained from direct otolith reading and those from back-calculation. Finally, the obtained results are compared with those from other areas.

Abstract reference: WSAgeP_Pattoura_23

Is otolith weight of European hake (*Merluccius merluccius*) a useful tool for the estimation of age?

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Estimating the age of hake by using annual rings remains an expensive and time consuming method. The present study is investigating the opportunity of using otolith weight (OW) to estimate the age of hake. A total of 282 specimens were collected from 2 areas within DCF Program in 2013 with total length (TL) ranging from 82-730mm. From each specimen, sagittal otoliths were extracted, cleaned, photographed, weighted to the nearest 0.0001 g and stored dry. Ageing was based on the identification of annuli on the images of the right otolith using Image analysis and taking as date of birth the first of January. No statistical differences were found between left and right otolith weight, thus the right otolith was used for the analyses. OW was examined to identify relationships with the factors age, sex, maturity and study area and the continuous variables TL and otolith radius (R) using general linear models. OW was found to be statistically significant related only with age, sex, TL and R, thus all analyses were performed for each sex separately. The relationships of OW-TL and OW-R were found to be multiplicative in all cases; their slopes differed significantly between males and females. ANOVA analysis and LSD test showed that the mean otolith weight by age group differed significantly among them for both sexes. The relationship of OW-Age was linear for both sexes; no differences were detected for slopes and intercepts. These models were used to re-estimate the age of the examined individuals based on OW. The appropriate criteria (sensitivity, specificity and accuracy) were used to evaluate the classification performance of the OW-age model. OW was found as a useful tool for the estimation and/or validation of hake ageing, based on the results of the model between the otolith based age groups and the estimated ones.

Abstract reference: WSAgeP_Rey_04

Enhancing otolith microstructure by a plastic inclusion technique

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Daily growth increments (DGI) throughout juvenile and adult otoliths only show up after polishing to thin slices around 100 μ . Otoliths are intricate three-dimensional structures formed by aragonite crystals arranged in different directions causing frequent unreadable areas for age estimation. Despite this structural complexity, some practical techniques can enhance DGI observation to improve age interpretations. A plastic inclusion method widely used in bony parts of other vertebrates, allow filling the otoliths' interstitial spaces with polymethyl metacrilate before the process of embedding in resin. As a result, plastic inclusion causes the structural reinforcement of the otolith permitting thinner and harder slices than those obtained from the standard process, improving DGI identification, even if it implies a moderate time-consuming process. A total 20 pairs

of otoliths from black hake otoliths (*Merluccius polli* and *M. senegalensis*) have been analyzed, comparing left (plastic inclusion process) and right (common process) otolith images contrast.

Abstract reference: WSAgeP_Silm_06

Estimating the age and growth of European eel (*Anguilla anguilla*) in Estonian lakes using „burning and cracking“ method on otoliths

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The eel population in the Narva River basin is based entirely on stocking. Prior to year 2001 eels were stocked into lake Võrtsjärv (270 km²) mostly in glass eel stage while from 2001 to 2010 only as elvers. From year 2002 young farmed eels were introduced to smaller lakes such as Lake Kaiavere, Lake Vagula, Lake Saadjärv and after the year 2003 Lake Kuremaa. More than 900 eel otoliths were gathered from 1999 to 2012 and treated with „burning and cracking“ method. In Lake Võrtsjärv the average annual growth rate of the eel stocked in glass eel stage was faster by 0,6 cm a year compared to the eels released as elvers, although the latter were 8 cm longer at the time of release. General growth rate was very good – 6,7 cm per year, being higher than indicated by previous results obtained in 1998 (5,9 cm). In four smaller waterbodies it was found that most likely the age of eel was overestimated due to the deposition of supernumerary zones in a rapid growth period, being slower from Lake Võrtsjärv by 3,4 cm per year.

Abstract reference: WSAgeP_Sion_14

Otolith ageing and preliminary validation of *Lampanyctus crocodilus* (Risso, 1810) in the North-western Ionian sea (Central Mediterranean)

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The jewel lanternfish, *Lampanyctus crocodilus* (Risso, 1810), is a mesopelagic fish living in the Eastern Atlantic and in the Mediterranean Sea where it is distributed within 1000 m in depth. Up to date, the knowledge on the bio-ecology of *L. crocodilus* is rather scarce and the few available regards taxonomic, trophic features or geographic and bathymetrical distributions. In this paper the study on age and growth of this myctophid fish in the Central Mediterranean is presented using direct and indirect methods. Samples were collected during experimental trawl surveys carried out from 2000 to 2004 in the North-western Ionian Sea as part of the MEDITS project. A total of 924 individuals were collected and 436 otoliths were analyzed. The standard length (SL) was measured and the maximum age of 8 years was assigned. The length at age key (ALK) was estimated by means of otolith readings and the von Bertalanffy growth parameters were estimated as well as the growth performance index (Φ') for the whole sampled population: $L_{\infty} = 231 \pm 17.45$ mm SL; $k = 0.11 \pm 0.015$ /year; $t_0 = -1.44 \pm 0.14$; $\Phi' = 3.77$. The results of ALK were compared with the modal component (Bhattacharya method) of length-frequency distribution selected during 2002 where the

greater number of individuals belonging to older age classes were sampled. The differences between the two methods, evaluated by means of Kruskal-Wallis rank test, resulted not statistically significant ($p > 0.05$). Moreover, the otolith reading was corroborated by back-calculation carried out according the Fraser-Lee method). Indeed the statistical comparison of the growth curve calculated by otolith reading and back-calculation were not significantly different.

Abstract reference: WSAgeP_Smith_09

Age Validation of lemon sole (*Microstomus kitt*), using marginal increment analysis

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Age estimation of the lemon sole (*Microstomus kitt*), a commercially valuable flatfish, has traditionally been estimated from otoliths using the 'breaking and burning' technique, however, perceived annual incremental growth has never been validated for this species. Here, the otoliths from 838 lemon sole collected from the North Sea, English Channel and Celtic Sea throughout entire months of 2009 were analysed. The periodicity of increment formation was examined using marginal increment analyses (MIA). No incremental growth was observed between January and March for any size class. Incremental growth generally commenced earlier in younger fish (April in ages 3-5) and later in older fish (May for ages 6-10, June for ages 11+). The results clearly demonstrate that a single annulus represents one year of growth. These results inform best practice, and provide greater confidence in age estimation and consequently, the quality of data used in the assessment of the stock.

Abstract reference: WSAgeP_Valero_21

Description of the main sources of error associate with the age allocation of Chilean jack mackerel *Trachurus murphy* (Nichols, 1920)

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In Chile since 1981 an annual periodicity in the band of annual growth for *Trachurus murphy* has been identified. This is consistent with estimates of the *Trachurus* genus but their identification is not an easy task and not conclusive. The first annual ring and absolute age for *T. murphy* was validated through radiocarbon method. However uncertainty remains in the interpretation of the first three growth annuli because false rings can be easily confused with 2nd or 3rd annulus. This work aims to define the qualitative and quantitative characteristics for the false and true annuli during the first three years of the fish growth. Readings of 90 otoliths pairs from three trained readers were compared. Growth analysis of the otolith radius between the first, second and third ring was performed. The age distribution estimation between readers was similar in a 46% of overall agreement percentage and 93% if the difference between the samples with ± 1 annulus was considered. We identify the area close to the nucleus where the highest presence of false rings was located at 2.8 and 3.2 mm from the core to the postrostrum. This area showed the largest difference of agreement in the reading criteria. Irregular annual growth marks between the second

and third growth ring could be related to ontological changes and evidence of onset of sexual maturity, with low somatic growth (allometric), producing disturbances in this area of the otolith. We propose a protocol for readers based on validation and in the consensus of criteria between the expert readers and bibliographic background. This protocol contains the following steps: 1) Observation external side 2) Observe the otolith edge 3) Check marks in the rostral zone 4) look inner face 5) analysis of marginal increment radius between the first to third ring.

Otolith Shape Analysis Workshop

Time		Moderator	Presenter	Title
09:00		Audrey Geffen	Audrey Geffen	<i>Introduction to otolith shape analysis; theory behind the practice</i>
09:10	WSShape Worsøe Clausen_01		Lotte Worsøe Clausen	Herring otolith shape – a tool for gauging stock complexity?
09:30	WSShape Vignon_04		Matthias Vignon	Disentangling and quantifying sources of otolith shape variation across multiple scales using a new hierarchal partitioning approach
09:40	WSShape Rivera__02		Veronica Rivera	Morphological analysis applied to the shape of the sagitta of <i>Fundulus persimilis</i> (Cyprinodontidae).
09:45	WSShape Rubio_03		Jacob Rubio	Morphometric analysis of the sagitta otoliths of <i>Pterois volitans</i>
09:50	WSShape Megalofonou_06		Persefoni Megalofonou	Otolith growth and shape changes during ontogeny in Atlantic bluefin tuna
09:55	WSShape Baudouin_07	Lotte Worsøe Clausen	Marie Baudouin	Discriminating stocks of common dentex (<i>Dentex dentex</i>) around Corsica Island (NW Mediterranean) using two otolith shape classification methods
09:55	WSShape Clausen_08		Lotte Worsøe Clausen and Antoni Lombarte	Image acquisition - code of best practice for obtaining high quality images with unambiguous outlines
10:40	Break			
11:00	WSShape Brophy_09	Deirdre Brophy	Deirdre Brophy	Image processing, outline generation and shape data extraction; interactive exercise
12:10	WSShape Libungan_11		Lisa Anne Libungan	shapeR: an R package to study otolith shape variation
12:30			Deirdre Brophy	What next? Introduction to analysing and interpreting otolith shape variation
13:00	Lunch			
14:00	WSShape Harbitz_12	Alf Harbitz	Alf Harbitz	Assessment and avoidance of pitfalls by use of Fourier techniques in discrimination analysis of otolith contours from different stocks; interactive exercise
15:05			Henrik Mosegaard	Statistical analysis of population assignment for fisheries management employing otolith characteristics; interactive exercise
15:45	Break			

Time	Moderator	Presenter	Title
16:05	WSShape Mosegaard_13	Henrik Mosegaard	(CTD) Statistical analysis of population assignment for fisheries management employing otolith characteristics; interactive exercise
16:30	WSShape ElHabouz__05	Deirdre Brophy	Youssef El Habouz
16:40	WSShape Harbitz_14	Alf Harbitz	Otolith identification system based on image contour analys A conceptual modification of three Fourier techniques to represent 2D closed contours that requires only one frequency to reproduce a pure ellipse
16:50	WSShape Marti-Puig_10	Pere Marti- Puig	Parameterization of 3D AFORO otolith surfaces
17:00	WSShape Fisher_15	Mark Fisher	Utility of Computer Assisted Age and Growth Estimation (CAAGE) in the analysis of otolith images for separating herring spawning groups.
17:10	WSShape Zitek_16	Andreas Zitek	Affordable 3D scanning of small otoliths for improved shape analysis by photogrammetry techniques
17:10	WSShape Loh_17	Kar-Hoe Loh	Morphometric analysis of the sagitta otoliths of Sciaenidae from Malaysia
17:10			<i>Panel Discussion</i>
18:00	End		

Otolith Shape Analysis Workshop Oral Presentations

Abstract reference: WSShape_Worsøe Clausen_01

Herring otolith shape – a tool for gauging stock complexity?

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Both genetic and environmental influences have been reported as important in determining the shape of the otolith. Different genotypes potentially induce important differences in otolith shape. Likewise, environmental differences between local habitats give rise to variation in the shape of otoliths. Otolith shape is currently used in stock assessment to separate mixed catches of the two major herring stocks in their common summer feeding grounds in the Eastern North Sea, Skagerrak and Kattegat. Herring are classified as either belonging to the autumn spawning North Sea stock or the spring spawning Western Baltic stock. However, several genetically differentiated populations have been identified within the Western Baltic stock. In this study, otolith shape was examined in samples of Western Baltic spring spawning herring collected both on spawning locations in the Western Baltic Sea and on mixed feeding grounds in the Skagerrak. Otolith shape variation in the mixed feeding aggregations was compared to the variation within and among the selected spawning aggregations to analyse the stock complexity in the Western Baltic herring. We tested whether otolith shape parameters corresponded with genetic marker based population identification of individuals in the mixed stocks, and whether shape based estimates of sample composition may be a proxy for population complexity. Based on these results, the application of otolith shape as a population identification tool for discrimination between genetically differentiated populations spawning in the same season is discussed.

Abstract reference: WSShape_Vignon_04

Disentangling and quantifying sources of otolith shape variation across multiple scales using a new hierarchical partitioning approach

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Otoliths have long been used for annual age and growth estimates but it have been increasing interest in the use of otolith shape as a natural tag of fish stocks because it provides a practical basis for stock separation that is useful in fisheries management. It is indeed currently recognised that wide range of environmental and genetic factors lead to otolith shape variation and the challenge for the future of morphometric stock identification is to develop a consensus on biological interpretations of otolith shape variation. Since otoliths have received considerable attention, it is fundamental to identify and quantify the relative contribution of the main factors that affect their shape variability. However, to date, there are few comprehensive reports that have examined simultaneously these different factors at multiple scales in natural conditions. In this study, I

disentangled sources of otolith shape variation across hierarchical spatial and taxonomic scales. Specifically, a comparative, morphometric study was made based on thousands sagitta otoliths from 8 species (Serranidae and Lutjanidae exclusively) in the Pacific Ocean. The effect of environmental, intrinsic and taxonomic factors on otolith shape variability was investigated, reflecting the likelihood of factor that one can detect when otolith shape is affected by multiples components. For this purpose, I propose a new hierarchical partitioning method embedded in a geometric morphometric framework. While large-scale variations are expected to be associated with important shape variation, this study provides the first quantitative demonstration that local environmental variables contributes equally to total shape variation than large-scale patterns. More generally, understanding of the scaling of otolith shape variation is critical for the use of otolith morphology as an effective tool to improve our understanding of the integrity of fish populations and the management of fisheries resources.

Abstract reference: WSShape_Rivera_02

Morphological analysis applied to the shape of the sagitta of *Fundulus persimilis* (Cyprinodontidae).

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The shape of otoliths provides information to discriminate fish stocks, populations, and species. Shape is also known to change along ontogeny and, in some species, among sexes. We used the geometric analysis of shape to study the sagittae of *Fundulus persimilis*, an endemic species of the northern coast of Yucatan, Mexico, which is subject to special protection according to a recent addendum to the environmental law. There is little biological information on this species and no contributions on the effect of individual growth on the shape of its otolith have been published. In this work, we describe the shape using 50 semilandmarks placed in the contour of the *sagittae* of individuals belonging to three size classes. Results of the canonical variates analysis (CVA) showed a clear separation between the shapes of otoliths of the three groups, there were significant differences between them (*Godall's F-test* $p < 0.05$). Morphological changes during growth are important in the posterior-dorsal and the anterior part of the otolith, mainly in the notch and the *antirostrum*. Results of the CVA and *Godall's F* value (3.61; $p < 0.05$) indicate that otoliths of males and females have different shapes. Fish and otolith size explain 80% and 61% of the otolith shape variation. The unexplained variance could be attributed to changes in abiotic factors of the highly dynamic habitat where this species live.

Abstract reference: WSShape_Rubio_03

Morphometric analysis of the sagitta otoliths of *Pterois volitans*

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Geometric morphometrics has proven to be a useful tool to investigate shape. In this work we evaluated the effect of ontogeny and external coloration on the shape of both left and right sagittae of the Lionfish (*Pterois volitans*), to group organisms with similar characteristics. Fish were obtained, photographed, and measured (SL, mm \pm 1) at a location in the coral reef system off the coast of the Mexican Caribbean. Otoliths were removed by dissection of the head of the fish and only complete pairs of sagittae were used to ensure comparable results. A total of 110 pairs of otoliths from fish in the range 81 mm \leq SL \leq 337 mm were photographed and shape was analyzed by placing 50 semilandmarks in the contour of the sagittae of individuals belonging to eight size classes, considering the three external color patterns (light, medium, dark). CVA results show that the shape of right otoliths are significant among all size classes of fish while left otoliths of only three size classes showed differences. Changes in the morphology of otoliths are particularly evident in the sulcus acusticus, which is better defined in late ontogenic stages where the cauda becomes deeper and cristas begin to appear in its terminal section. As size increases, the notch becomes progressively conspicuous but it does not appear in all organisms. The posterior part of the otolith is the most variable region, its shape ranging from pointed to rounded, and even truncated, presenting irregular undulations along the edge.

Abstract reference: WSShape_Megalofonou_06

Otolith growth and shape changes during ontogeny in Atlantic bluefin tuna

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The otoliths grow over the entire life time of a fish. Both genetic and environment factors contribute to the variation observed in growth rate and shape of otoliths while it is thought that these are controlled by the protein synthesis and growth of fish. The main objective of this work was to study the growth of sagittal otoliths in Atlantic bluefin tuna and examine the changes of otolith shape in relation to fish size. The hypothesis that otolith size or shape variables are directly related to fish age is also considered, with the aim of providing strictly objective measurements that could be used as valuable criterion to estimate age of Atlantic bluefin tuna. Otolith morphological characteristics were studied in specimens ranging in fork length from 8.5 to 278 cm and from 20 days to 21 years in age. Four morphometric variables - whole otolith length, width, area and perimeter - were measured using image analysis techniques and three shape indices - circularity, E value and rectangularity - were calculated for each pair of sagittal otoliths. The absolute and relative growth of otoliths was determined. Mean otolith lengths at age were predicted from the otolith growth equations. Statistically significant relationships were observed between all otolith variables tested and the age or length of fish. Among the variables, otolith area was the one that showed the

highest correlation with fish length ($R^2=0.983$, $Ao=0.141FL1.136$) followed by otolith length ($R^2=0.978$, $Lo=0.541FL0.654$) whereas the otolith circularity exhibited the lowest correlation. Remarkable changes in otolith shape indexes were observed during the early stages of bluefin tuna ontogeny.

Abstract reference: WSShape_Baudouin_07

Discriminating stocks of common dentex (*Dentex dentex*) around Corsica Island (NW Mediterranean) using two otolith shape classification methods

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The common dentex *Dentex dentex* (Linnaeus, 1758) is a Mediterranean and near Atlantic coastal top predator fish. This species is of great economic importance for both artisanal (small-scale coastal fisheries) and recreational fishing. The common dentex is also the only sparid fish classified by the International Union for the Conservation of Nature (IUCN) as "Vulnerable" in the Red List of Threatened Species in the Mediterranean Sea. Despite its ecological and economic importance, data on the stock structure of this species are still very scarce. The aim of this study was to determine the spatial structure of common dentex around Corsica Island using otolith shape. The shape analysis has often been successfully applied using shape index and elliptic Fourier analysis. In this study we also used geometric morphometrics, a less common method that appears slightly more efficient for visualization purpose and can therefore be used as a useful complementary method for studying otoliths shape variations. For this purpose 95 otoliths were collected from four different zones around Corsica. First, multiple regressions were applied to test the influence of endogenic variables (size, weight and gender). It appears that these variables contribute significantly to otolith shape variations, as well as the geographic location. Canonical analysis shows a spatial pattern, particularly for the region of Ajaccio which is well-segregated. However the sample region explained only 0.5% of the total shape variation, emphasizing that other environmental factors may affect otolith shape variations among individuals. The use of the two complementary methods (*i.e.* Fourier series and geometric morphometrics) is discussed. Nevertheless, these first results support the use of otoliths shape as a tool for understanding spatial population structure of this species.

Abstract reference: WSShape_Worsøe Clausen_08

Image Acquisition – do's, don'ts, and important things to remember Lotte Worsøe Clausen¹ and Antoni Lombarte²

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Otolith shape analysis is a widely used technique for e.g. species- or stock identification and separation. There are several steps in shape analysis which each are equally important for the validity, ease and efficiency of the result from any analyses of otolith shape as they all potentially

can bias the perception of the actual otolith shape. Here we demonstrate how the image acquisition process can introduce artefacts if not carefully considered prior to any analysis of otolith shape. The steps involved in acquiring standardised images for otolith shape analysis are demonstrated with particular focus on contour acquisition by light microscope of the whole otolith and sulcus acusticus (internal side) shapes of otoliths. Potential caveats in relation to the physical otolith will be identified (e.g. anatomical position, side asymmetries, crystallizations anomalies, broken tips, otolith rotation in relation to curvature) as well as important factors to consider in relation to the image acquisition set-up (e.g. light settings, background and medium) and approaches to dealing with this error will be outlined. Additionally a demonstration of methods for capture of internal otolith shape will be made along with guidance as to how to identify the optimal structures for analysis. The session will be a combination of presentations and live demonstration of image acquisition and image digital processing illustrating a series of issues to take into consideration when acquiring photos of otoliths. The aim is for participants to leave with an outline of what to include when setting up a procedure for otolith shape acquisition minimising the potential bias of image artefacts on the following otolith shape descriptor extractions.

Abstract reference: WSShape_Brophy_09

Obtaining numerical shape descriptors from otolith images.

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Otolith shape analysis is a relatively inexpensive technique that can be readily applied using widely available microscopy and digital imaging equipment coupled with freely available software tools. Rigorous application of the technique to questions of stock structure requires that shape descriptors are independent of size and are not subject to artefacts of image acquisition. A conceptual explanation of commonly used shape descriptors (basic shape indices, elliptical fourier descriptors (EFDs), landmarks, wavelets) will be presented. The steps involved in obtaining shape data (primarily focusing on EFDs) from images will be outlined (obtaining outline; extracting EFDs; smoothing; deciding on number of harmonics; standardising for size, orientation, starting point). Sources of potential error/noise will be identified (e.g. ambiguities in the outline; image rotation; size/magnification) and approaches to dealing with this error will be outlined. Available software resources for conducting shape analysis will be identified and analytical methods for removing size or age effects from shape descriptors and comparing or identifying groups will be introduced. Tools for visualising group differences will also be presented. The overall aim of the session is to equip workshop participants with the skills and software tools needed to generate outlines and extract shape descriptors from otolith images, to inform them of key methodological limitations and how to deal with them and to introduce them to available approaches to analysing and interpreting otolith shape data.

Abstract reference: WSShape_Libungan_11

shapeR: an R package to study otolith shape variation

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shapeR is an open source software package that runs on the R platform for the collection and analysis of otolith shape data. The package allows comparison among populations of fish within the same species sampled at small and large geographic scales. The method might be used as well for species comparison and for analyses of variation in shape in two dimensions. shapeR reads images into R, collects the outlines of the otoliths, performs a Discrete Wavelet transform on radiuses and a Normalized Elliptic Fourier transform on the otolith outlines. Comparison of the multiple independent Wavelet or the Fourier coefficients within and among populations are done with multivariate statistical analysis.

The Wavelet transform provides a powerful alternative to the Fourier transform in shape analysis. Fourier transform is a useful tool when overall differences in otolith shape need to be analyzed, however it fails when more detailed information of the shape differences are needed, for example to evaluate which areas of the otolith outline are contributing most towards the variation among populations. The advantage of using Wavelet over Fourier in shape analysis will be discussed and examples will be shown by applying the shapeR package; including how to perform quality checks when collecting otolith outlines, how to visualize the mean shape of each population and how to apply statistical tests to the output of the package.

Abstract reference: WSShape_Harbitz_12

Assessment and avoidance of pitfalls by use of Fourier techniques in discrimination analysis of otolith contours from different stocks

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The attention is restricted to the linear Fisher discrimination technique applied to Fourier coefficients of standardized otolith contours from different stocks, and use of the “leave one otolith out at a time” technique to obtain reliable score estimates. Two pitfall categories are considered: Covariates that are not taken into account, and technical artifacts related to image analysis. A typical example of the former category is age: Two samples with equal size and sex distribution may appear to have different shapes due to different age distributions. An example of a technical artifact is otolith orientation, because the pixel noise is influenced by the orientation of the otolith, even though the contour is standardized to the same orientation by rotation. The effect of the latter can be assessed by rotating the original images before extracting the contour and then examine if the sample of otoliths with and without rotation are separated. A general theoretical result that a discrimination score equal to zero is always obtained when comparing two identical samples, is shown, along with examples of how this result can be applied to assess the impact of technical

artifacts. As a tool to diminish the effect of artifacts, the concept of a lasso contour is introduced that consists of straight lines between the pixel points that constitute a non-concave contour shape. It appears that the lasso contour diminishes the effect of pixel noise and rotation, and is well suited to the 1D mirror (reflection) technique, because ambiguities are avoided. Results are illustrated with Greenland halibut and cod otolith samples.

Abstract reference: WSShape_ Mosegaard_13

Statistical analysis of population assignment for fisheries management employing otolith characteristics

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Traditionally fisheries management is area based and single stock oriented. However biological populations do not always adhere to fixed boundaries. Many fish stocks defined by managers have underlying complex population structures with diverging dynamics and overlapping distribution patterns. To maintain full productivity fisheries management should be based on the most unbiased assessment of individual population dynamics within a stock complex.

By their genetically and environmentally controlled growth pattern otoliths have gained increasing interest in quantitative fisheries biology. In this study we focus on how extracted information from otolith characteristics like shape and growth pattern may be analyzed to inform management decisions on exploitation opportunities in situations of population mixing.

Biological data from different important fish stocks each comprised of several populations are analyzed and used to develop synthetic populations with corresponding otolith and fish parameters for further Monte Carlo simulations of the population assignment process.

Given a suite of otolith related parameters, the preconditions for optimal baseline selection for population assignment in mixed stocks are analyzed. Different discrimination methods and bias corrections are compared in relation to their operational use in population dynamics. Different approaches are discussed dependent on whether the aim is unbiased proportions of population affiliation at age or precise individual based life history parameters like size or maturity at age.

Abstract reference: WSShape_ElHabouz_05

Otolith identification system based on image contour analysis

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In this paper we present an automatic classification system of fish species based on otolith contour analysis. Otoliths have a distinctive external form which is usually a characteristic of the species. The external shape of the Otolith varies according to species, but it's substantially constant in the same species. This can be used as a characteristic of fish species recognition. Identification of fish species using Otoliths is a major issue in many marine ecological studies. For example, the Otoliths

recovered from the stomach or feces could be used to determine the food spectrum. The proposed system consists of three main phases: pre-processing, feature extraction and classification. The first phase is the image denoising and enhancing grayscale contour to facilitate the detection and contour extraction. In the second phase, we extract the median distance vector of the contour. This vector is used in Otolith recognition phase, which is based on the Support Vector Machine (SVM) classification method. The system was tested on a set of 60 fish Otolith images from AFORO database, 10 images per species for six species (*Scomber colias*, *Coris julis*, *Umbrina canariensis*, *Diplodus annularis*, *Trachurus mediterraneus* and *Trisopterus minutus*). The experimental results show the robustness of the approach (98.33%).

Abstract reference: WSShape_Harbitz_14

A conceptual modification of three Fourier techniques to represent 2D closed contours that requires only one frequency to reproduce a pure ellipse

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Elliptical Fourier Descriptors (EFDs) is probably the most extensively applied technique in shape analysis of a closed contour, with a range of applications within discrimination between fish stocks based on EFDs of otolith contours. A recent technique transforms the 2D contour to a 1D function by mirroring (reflecting) the lower half of the contour around a vertical axis at the right end of the contour. This technique has the advantage of reducing the number of Fourier coefficients to two coefficients per frequency component compared to four with the EFD. The third technique considered is also a 1D technique, based on the tangent angle to the contour at equidistant distances along the contour. This technique also needs only two coefficients per frequency component. All techniques, however, require several frequency components to reproduce a pure ellipse properly. This is due to the applied concept of a constant speed along the contour in the EFD case, equidistant “time steps” in the 1D mirror technique and equidistant contour distance sampling in the 1D tangent angle technique. This contribution shows how these techniques can be easily modified by relaxing on the “equidistant” sampling approach, resulting in a virtually perfect reproduction of a pure ellipse with only one frequency component. In addition, examples are shown where the modification gives better approximation to the original contour with fewer coefficients than the original EFD and the 1D mirror techniques. Results are demonstrated on Greenland halibut as well as cod otolith samples.

Abstract reference: WSShape_Marti-Puig_10

Parameterization of 3D AFORO otolith surfaces

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The 3D otolith shapes recently included in the AFORO database are defined by means of clouds of

points of their 3D surfaces. Automatic retrieval and classification of 3D natural objects becomes a difficult task when the number of the elements in the database increases because of the great amount of information involved in the description of each object. In order to simplify that task, in this contribution we propose a new method for compacting data from 3D shapes by extracting, for each shape, a 3D characteristic curve that works as a 3D object signature. By means of that 3D signature the morphological shape information can be preserved but the number of points required for the shape representation is drastically reduced. The method allows us to define a 3D characteristic curve with a variable precision, so if we need 3D objects with a low precision the number of points diminishes also. The important issue is that, by construction, those signatures always form a 3D closed curve and their corresponding x, y, z coordinates can be treated as periodic functions. This property lets apply Elliptical Fourier descriptors on each coordinate in order to achieve a bigger information compression if we select a reduced set of them (the ones that bring more information). So, with a reduced set of parameters, the main information of the original 3D shape can be preserved. The 3D otolith shapes of AFORO are digitalized following an acquisition protocol and therefore it is not necessary that their descriptors have the rotation invariance property. However, from the proposed set of parameters, it is possible to develop a new set of parameters that are invariant to rotations. The presented parameterization was applied to sagitta otolith of Mediterranean sciaenids but can also be applied to describe other biological origin shapes.

Abstract Reference: WSShape_Fisher_15

Utility of Computer Assisted Age and Growth Estimation (CAAGE) in the analysis of otolith images for separating herring (*Clupea harengus*) spawning groups.

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Building on recent work reporting successes of fully automatic CAAGE systems applied in aging of commercially important demersal fish species this study investigates the feasibility of using the technology for stock discrimination. We review the typical imaging pipeline of CAAGE systems highlighting popular algorithms and potential pitfalls. We review morphological image processing techniques for identifying the otolith core and we compare signal processing algorithms for analysing 1-D transept profiles originating from the core and extending to its edge. We discuss practical problems that arise when applying these approaches to species of pelagic fish and show that the methods are sensitive to parameter selection and other factors associated with image acquisition. Then we discuss how systems that employ an interactive user interface overcome some of the problems encountered with fully automated systems, while still providing benefits in terms of improved efficiency. To illustrate this work we focus on a benchmark problem concerning the reassignment of Thames and North Sea herring individuals to their source populations by measurement of year 1 incremental growth. We perform interspecies classification experiments using linear discriminant analysis (LDA) on measurements made on otolith images by an expert reader, an automatic system, and an interactive system. Our fully automatic system achieves an accuracy of >70% while an expert working with conventional image annotation tools can achieve >85%. Our results demonstrate that an expert working interactively with a computer assisted age

and growth estimation system can quickly correct errors made by the system and improve efficiency.

Abstract reference: WSShape_Zitek_16

Affordable 3D scanning of small otoliths for improved shape analysis by photogrammetry techniques

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Until today 3D scanning of otoliths has been achieved with high-end instrumentation (Micro-CT) or only for otoliths larger than 1 cm in diameter e.g. by using a specialized projector and white light setup. So far, no affordable system for smaller otoliths (below 10 mm in diameter) exists, allowing for the archiving and accurate evaluation of otolith geometries.

Here, we present a new, comparably low-cost, system for 3D scanning of small otoliths (from 0.5 mm to 5 mm length), based on photogrammetry techniques. The hardware system consists of an USB microscope, an Arduino Uno Controller board, a stepping motor with a stepping motor driver and a self-designed and 3D printed stage. AgiSoft Photoscan is the underlying software system for the creation of the 3D model and works on the basis of merging sequentially recorded pictures. In total the system costs less than 600 U\$. Once a 3D model of the otolith is established, the otolith model can be virtually cut along various planes yielding different 2D sections that can be then subjected to shape analysis. Compared to projector and white light based 3D scanning, the presented system is also able to capture the natural texture of the scanned otolith which provides additional information for interpretation. The method presented is expected to significantly improve the possibilities for differentiating and classifying fish from different sub-populations.

Abstract reference: WSShape_Loh_17

Shape analysis of the sagittal otoliths of Sciaenidae from Malaysia

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Fish otoliths have been widely used for identification purpose, and there is a growing number of otolith image databases being established. Through outline analysis of sagittal otoliths, an identification system using geometric morphometrics was developed. The system provides automated routines for image processing, outline sampling, evaluation of classification models, as well as search and predict functions. The system was tested on the sagittal otoliths of Malaysian Sciaenidae, including 9 common species from 8 genera. Classification performance showed over 96% of overall accuracy, using either Procrustes analysis on sliding semi-landmarks or elliptical Fourier analysis. Besides analyzing the outline of the proximal aspect of the right otolith, outlines of other aspects (distal, anterior, posterior, dorsal, and ventral) were also tested. Proximal aspect has

the best classification performance compared to other aspects, but combination of different aspects slightly improved the performance. Although allometric relationships between shape and size of otolith was significant ($p < 0.001$) in some of the species such as *Panna microdon*, *Nidea soldado*, and *Otolithes ruber*, the ontogenetic changes does not affect the classification performance.