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SCICOM STEERING GROUP ON ECOSYSTEM PRESSURES AND IMPACTS

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Interim Report of the Stock Identification Methods Working Group (SIMWG)

By correspondence

International Council for the Exploration of the Sea

Conseil International pour l'Exploration de la Mer

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Contents

Executive summary

Over the past year, the Stock Identification Methods Working Group (SIMWG) has made progress toward addressing our multi-year terms of reference and has contributed to ICES Science Plan priorities. The working group was chaired by Lisa Kerr (USA) during this period and SIMWG worked by correspondence in 2017.

SIMWG has continued to provide annual updates on recent applications of stock identification methods to ICES species and on advances in stock identification methods. SIMWG's annual reviews on advances in stock identification methods keeps ICES members abreast of best practices in this field of study.

SIMWG continues to work on resolving issues of stock identification as requested by ICES working groups. Over the past year, we have received requests for advice on Atlantic sardine from participants in the Workshop on Atlantic Sardine (WKSAR) and a request from the North Western Working Group (NWWG) to review beaked redfish (*S. mentella*) stock affiliation on the East Greenland slope. These reviews are ongoing.

SIMWG continues to work on reviewing and reporting on advances in mixed stock analysis. This work is relevant to resolving mixed stock composition issues in assessment and management and will continue over the next two years of our three year cycle.

1 Administrative details

Working Group name

Stock Identification Methods Working Group (SIMWG)

Year of Appointment within current cycle

2017

Reporting year within current cycle (1, 2 or 3)

1

Chair(s)

Lisa Kerr, USA

Meeting venue and dates

By correspondence

2 Terms of Reference a) – z)

SIMWGs multiannual ToRs are as follows:

a) Review recent advances in stock identification methods;
b) Provide technical reviews and expert opinions on matters of stock identification, as requested by specific Working Groups and SCICOM;
c) Review and report on advances in mixed stock analysis, and assess their potential role in improving precision of stock assessment.

3 Summary of Work plan

Year 1	Address terms of reference through work by correspondence in 2017
Year 2	Organise a physical meeting for SIMWG for summer 2018.
Year 3	Address terms of reference through work by correspondence in 2019.

4 List of Outcomes and Achievements of the WG in this delivery period

- SIMWG provided an annual updates on recent applications of stock identification methods to ICES species and on advances in stock identification methods.
- SIMWG is planning an in-person working group meeting in 2018.

- SIMWG has received two requests for review of stock identification issues (Atlantic sardine and beaked redfish) and these reviews are currently underway and will reported on in the coming year.
- SIMWG continues to consider advances in mixed stock analysis and plans to dedicate effort during the coming year to outline a paper relevant to resolving mixed stock composition issues in assessment and management.

5 Progress report on ToRs and workplan

ToR a) Review recent advances in stock identification methods

In the last year, there was a proliferation of applications of stock identification methods to ICES stocks, as well as several notable advances in stock identification methods with many results relevant to ICES science and advice. SIMWG experts summarized new applications and advances with a focus on recent research in genetics, growth marks in calcified structures, life history parameters, morphometrics, tagging (conventional, acoustic, satellite), otolith shape, otolith chemistry, parasites, simulation approaches, and interdisciplinary approaches (see Annex 3 for review).

ToR b) Provide technical reviews and expert opinions on matters of stock identification, as requested by specific Working Groups and SCICOM

SIMWG has received two requests for review of stock identification issues and these reviews are currently underway and will reported on in the coming year.

SIMWG received a **request** from the participants of the Workshop on Atlantic Sardine (WKSAR). Participants from WKSAR requested a review of stock identity information on Atlantic sardine. The group summarized available stock structure information in the WKSAR report, and requested feedback from SIMWG on the document and input on the specific question of the appropriateness of stock structure scenarios they have put forth for consideration in the next benchmark assessment process.

SIMWG received a request from the North Western Working Group (NWWG) to review beaked redfish (*S. mentella*) stock affiliation on the East Greenland slope. In 2009, ICES reviewed the stock structure of beaked redfish, *Sebastes mentella* in the Irminger Sea and adjacent waters (WKREDS). They recognised that there are three biological stocks of *S. mentella* in the Irminger Sea and adjacent waters: 'Deep Pelagic'; 'Shallow Pelagic'; and 'Icelandic Slope'. This separation of the stocks did not include *S. mentella* on the Greenland continental slope. ICES decided that NWWG should conduct a separate assessment for *S. mentella* in subarea 14.b until further information was available to assign stock origin. Since 2009, further studies on stock structure and species separation have been conducted. Based on this new information NWWG recommended that the separation of *S. mentella* on the Icelandic and Greenlandic slopes be revised and the possibility of a joint assessment of *S. mentella* on the Icelandic and Greenlandic slopes be evaluated. NWWG requested SIMWG review of this issue of stock identification.

ToR c) Review and report on advances in mixed stock analysis, and assess their potential role in improving precision of stock assessment

SIMWG continues to consider advances in mixed stock analysis and plans to dedicate effort during the coming year to outline a possible paper that would be a focus of work at our upcoming in-person meeting planned for next year. The aim would be to develop a paper that is relevant to resolving mixed stock composition issues in assessment and management.

6 Revisions to the work plan and justification

There have not been any significant revisions to the SIMWG work plan this year.

7 Next meetings

We are in the process of planning our 2018 ICES SIMWG meeting to be held at the University of Iceland in Reykjavik, Iceland. We are currently working to resolve how many members would be available to attend and the best timing for this meeting. In the past, we have held our meetings in June for 2–3 days, depending on our ToRs. A key focus of this meeting will be dedicating time to ToR b) Provide technical reviews and expert opinions on matters of stock identification, as requested by specific Working Groups and SCICOM; and ToR c) Review and report on advances in mixed stock analysis, and assess their potential role in improving precision of stock assessment.

Annex 1: List of participants

NAME	ADDRESS	EMAIL
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Annex 2: Recommendations

SIMWG does not have any formal recommendations this year.

Annex 3: ToR a) Review recent advances in stock identification methods

In the last year, there have been several notable advances in stock identification methods and a proliferation of applications, with many results relevant to ICES science and advice. Here, we summarize advances and results accounting for research in genetics, life history parameters, growth marks in calcified structures, morphometrics, tagging, otoliths, parasites, simulation approaches, and interdisciplinary approaches.

i. *Genetic Analysis (Contributor: Stefano Mariani)*
ii. *Life history parameters (Contributor: Richard McBride)*
iii. *Body Morphometrics (Contributor: Zachary Whitener)*
iv. *Tagging (conventional, acoustic, satellite) (Contributors: Helene De Pontual (lead), Steve Cadrin)*
v. *Otolith Shape (Contributors: Kelig Mahe, Christoph Stransky, Helene De Pontual*
vi. *Otolith Chemistry (Contributors: Lisa Kerr and David Secor)*
vii. *Parasites (Contributor: Ken Mackenzie)*
viii. *Simulation approaches (Contributor: Lisa Kerr*
ix. *Interdisciplinary analysis (Contributor: All participants)*
x. *Published Theme Set Update (Contributor: Lisa Kerr):*

Genetic Analysis (Contributor: Stefano Mariani)

Since the analysis carried out in Mariani and Bekkevold (2014), we are continuing to monitor changes in the usage of genetic methods in fisheries stock identification on an annual basis. Here we report data on the most recent ten years of scientific output (Figure 1). Through 2015 microsatellites continued to be the most widely applied genetic method, however in 2016 the relative importance of SNPs appears to become more substantial. SNP studies now account for approximately one third of the total applications for fish pop structure identification. SIMWG will continue to track and summarize trends in genetic methods, in order to monitor the short-term changes in molecular marker usage in fisheries science.

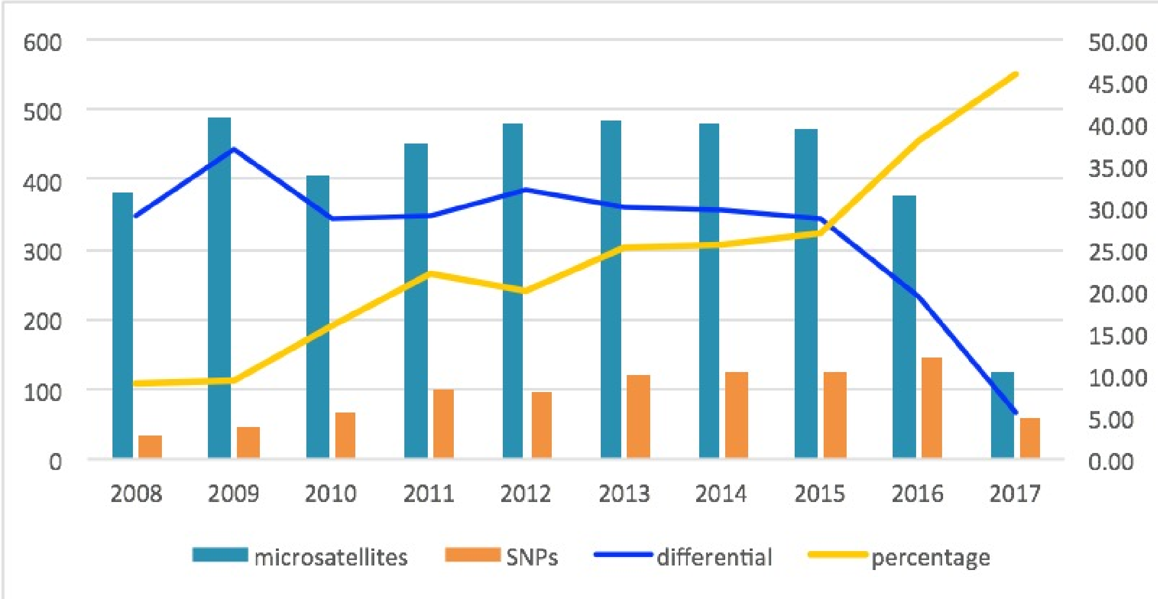

Figure 1. Scientific publishing trend since 2008, comparing outputs of studies using microsatellites (blue bars) and SNPs (orange bars), as listed in the ISI Thompson-Reuters Web-of-Science. The search criteria were: "fish* AND gene* AND (population OR stock) AND 'molecular marker*'," where 'molecular marker*' means "Microsatellite*" or "SNP*". Only papers in the following disciplinary areas were considered: 'Fisheries', 'Environmental Sciences & Ecology', 'Biodiversity Conservation', 'Marine & Freshwater Biology' and 'Oceanography'.

Below we summarize a few notable papers published in the past year:

Kennington *et al.* (2017) exemplifies the fact that many species that are commercially exploited and traded must still be assessed for structure, and that microsatellites are still useful and practical in many situations. Alfonso *et al.* (2017) shows that some related tasks, such as monitoring changes in effective population size, can be still be performed with low-coverage hypervariable markers. Martinez *et al.* (2017) and Jasonowicz *et al.* (2017) show that in many cases SNPs will not necessarily unveil new strong structure, compared to microsatellites. These papers demonstrate examples of species for which the application of genomic SNPs simply does not detect any meaningful structure.

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Jasonowicz, Andrew J.; Goetz, Frederick W.; Goetz, Giles W.; *et al.* 2017. Love the one you're with: genomic evidence of panmixia in the sablefish (Anoplopoma fimbria). Canadian Journal of Fisheries and Aquatic Sciences. 74(3): 377-387

Kennington, W. Jason; Keron, Peter W.; Harvey, Euan S.; *et al.* 2017High intra-ocean, but limited inter-ocean genetic connectivity in populations of the deep-water oblique-banded snapper Pristipomoides zonatus (Pisces: Lutjanidae). Fisheries Research. 193:242-249

Martinez, Edith; Buonaccorsi, Vincent; Hyde, John R.; *et al.* 2017. Population genomics reveals high gene flow in grass rockfish (Sebastes rastrelliger).Marine Genomics. 33:57-63

Pita, Alfonso; Perez, Montse; Velasco, Francisco; *et al.* 2017. Trends of the genetic effective population size in the Southern stock of the European hake. Fisheries Research. 191. 108-119.

Life history parameters (Contributor: Richard McBride)

An interdisciplinary approach to stock structure, with a nice balance of life history and genetic information, is emerging for European flounder (*Platichthys flesus*) in the Baltic Sea. Erlandsson *et al.* (2017) report that size, growth, and maturity vary in a clinal pattern in the Baltic (ICES subdivisions 25-28), suggesting that flounder in the Baltic consists of several loosely defined sub-populations. Such spatially-explicit phenotypic data are consistent with genetic data. In addition, differences in spawning patterns – demersal spawning in the low salinity regions of the northern Baltic versus pelagic spawning elsewhere – coupled with genomic divergence actually indicates emerging speciation in this region (Momigliano *et al.* 2017).

Large data sets of fish size and age, common to data-rich fisheries, can be exploited to learn more about stock structure. Barrios *et al.* (2017) report that growth of whiting (*Merlangius merlangus*) varied among most ICES divisions in the eastern North Atlantic. They examined growth trajectories of 100s of individuals, and length-at-age data of 1000s of individuals, using a mixed effects model. Although phenotypic traits such as growth may be environmentally driven, rather than genetically based, spatial variation in growth rates may affect fishery yield, and is therefore of interest to assessment modellers and managers.

References

Barrios, A., B. Ernande, K. Mahe, V. Trenkel, and M. J. Rochet. 2017. Utility of mixed effects models to inform the stock structure of whiting in the Northeast Atlantic Ocean. *Fisheries Research*, 190:132-139.

Erlandsson, J., O. Ostman, A. B. Florin, and Z. Pekcan-Hekim. 2017. Spatial structure of body size of European flounder (*Platichthys flesus* L.) in the Baltic Sea. *Fisheries Research*, 189:1-9.

Momigliano, P., H. Jokinen, A. Fraimout, A.-B. Florin, A. Norkko, and J. Merilä. 2017. Extraordinarily rapid speciation in a marine fish. *Proceedings of the National Academy of Sciences*, 114(23):6074-6079.

Morphometrics and Meristics (Contributor: Zachary Whitener)

Body morphometrics and meristics are important methods for stock discrimination and their use is well established. Although basic meristics have long been described for many species, for others, basic meristics at the taxonomic identification are still being established. These may be useful for stock identification purposes in the future.

Sley *et al.* (2016) describe the morphometric and meristic characteristics of two Carangidae species in the Gulf of Tunisia, the blue runner *Caranx cryso* and the false scad *Caranx rhonchus*. Although stock identification was not performed in this study, the authors provided baseline information that contributes to species identification in a multispecies fishery and this information may be useful for stock identification in the future.

Allaya *et al.* (2016) used 13 morphometric and 4 meristic characteristics to discriminate between Atlantic chub mackerel *Scomber colias* samples taken from 3 sites on the Tunisian coast. They were able to do so with an 83.87% and 49% correct classification rate, respectively. Differences between sample sites were hypothesized to be due to genetic or environmental differences.

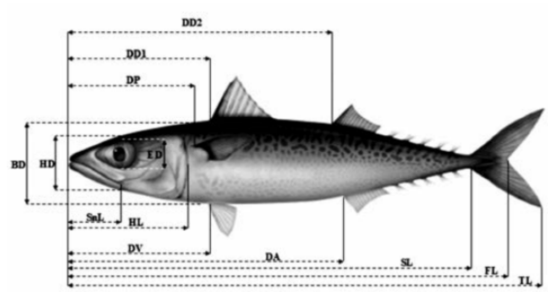

Figure 2. *Scomber colias.* Schematic drawing body with measured dimensions: (1) Snout Length (SnL), (2) Eye Diameter (ED), (3) Body depth (BD), (4) Head depth (HD) , (5) Head Length (HL), (6) Distance of pectoral fin (DP), (7) Distance of the first dorsal fin (DD1), (8) Distance of the second dorsal fin (DD2), (9) Distance of ventral fin (DV), (10) Distance of anal fin (DA), (11) Standard Length (SL), (12) Fork Length (FL) and (13) Total Length (TL).

Figure from Allaya *et al.* (2016).

References

Allaya, H., A. Ben Faleh, M. Rebaya, S. Zrelli, G. Hajjej, A. Hattour, J.-P. Quignard, M. Trabelsi. 2016. Identification of Atlantic chub mackerel *Scomber colias* population through the analysis of body shape in Tunisian waters. *Cahiers de Biologie Marine*, 57: 197-207.

Sley, A., L. A. Jawad, G. Hajjej, O. Jarboui, A. Bouain. 2016. Morphometric and meristic characters of blue runner *Caranx crysos* and false scad *Caranx rhonchus* (Pisces: Carangidae) from the Gulf of Gabes, Tunisia, Eastern Mediterranean. *Cahiers de Biologie Marine*, 57: 309-316.

Tagging (conventional, acoustic, satellite)
(Contributors: Hélène de Pontual, Steve Cadrin, Karin Hüssy)

Site fidelity and connectivity between largely remoted habitats was demonstrated on whale shark (Hearn *et al.*, 2016). An integrated assessment framework (IAF) was designed for understanding hammerhead population structure and connectivity where genetic and tagging data were used to produce conceptual models of stock structure and movement, generating several hypotheses (Chin *et al.*, 2017). Pop-up satellite archival tagging (PSAT) was used to investigate the survival, movements, and habitat use of mature female school shark as bycatch of demersal longlines targeting gummy shark in the Great Australian Bight (Rogers *et al.*, 2017). Hazen *et al.* (2016) used 16 years of electronic tagging data on bluefin tuna to predict the impact of oil exposure on spawning grounds of the Gulf of Mexico population. Below we report the abstracts of the cited papers.

Chin *et al.* (2017) studied population structure and connectivity of hammerhead shark species in the Australasian region. They developed an Integrated Assessment Framework capable of incorporating a suite of data, including a combination of genetic and tagging data. This study found geographic trends in stock structuring related to sex and size of the sharks.

Hazen *et al.* (2016) used electronic tagging data to identify habitat preferences of the heavily exploited bluefin tuna in the Gulf of Mexico, with the objective to evaluate the species' exposure to the Deepwater Horizon oil spill. The overlap between oil spill and spawning habitat was found to be limited, but combined with other drivers such as ocean warming and fishing mortality, may contribute to stagnation in stock recovery.

Hearn *et al.* (2016) tagged adult whale shark with satellite tags in the Galapagos Island with the objective to study stock structuring. They found strong temporal patterns in migration directions. Apparent return migrations documented a strong connectivity between populations at Galapagos and mainland Ecuador/Peru.

Using pop-up satellite tags, Rogers *et al.* (2017) studied survival, movements and habitat use of school shark in southern Australia after capture as bycatch by demersal automatic longlines with subsequent release. This study showed that school shark survive being released, exhibit a semi-pelagic habitat use, and undertake extensive offshore migrations.

References

Chin, A., Simpfendorfer, C. A., White, W. T., Johnson, G. J., McAuley, R. B., Heupel, MR. 2017. Crossing lines: a multidisciplinary framework for assessing connectivity of hammerhead sharks across jurisdictional boundaries. *Scientific Reports*, 7.

Hazen, E. L., Carlisle, A. B., Wilson, S. G., Ganong, J. E., Castleton, M. R., Schallert, R. J., Stokesbury, M. J. W., *et al.* 2016. Quantifying overlap between the Deepwater Horizon oil spill and predicted bluefin tuna spawning habitat in the Gulf of Mexico. *Scientific Reports*, 6.

Hearn, A. R., Green, J., Roman, M. H., Acuna-Marrero, D., Espinoza, E., Klimley, A. P. 2016. Adult female whale sharks make long-distance movements past Darwin Island (Galapagos, Ecuador) in the Eastern Tropical Pacific. *Marine Biology*, 163: 214.

Rogers, P. J., Knuckey, I., Hudson, R. J., Lowther, A. D., Guida, L. 2017. Post-release survival, movement, and habitat use of school shark Galeorhinus galeus in the Great Australian Bight, southern Australia. *Fisheries Research*, 187: 188-198.

Otolith Shape (Contributors: Kelig Mahe, Christoph Stransky and Helene De Pontual)

In the past year, there were 13 papers published using otolith shape analysis. Three of these studies detailed methodological developments and their applications to otolith shape analysis (Harbitz, 2016; Wong *et al.*, 2016; Mapp *et al.*, 2017) and one study compared otolith and scale shape (Ibanez *et al.*, 2017). Mille *et al.* (2016) observed the effect of diet on otolith shape. Additionally, eight papers used otolith shape as a tool for stock identification (round sardinella *Sardinella aurita,* Bacha *et al.*, 2016; vocal toadfish *Porichthys notatus,* Bose *et al.*, 2017; sharpsnout seabream *Diplodus puntazzo,* Bostanci *et al.*, 2016; *Atherina boyeri*, Boudinar *et al.*, 2016; *Mugil cephalus,* Callicó Fortunato *et al.*, 2017a; *Mugil liza,* Callicó Fortunato *et al.*, 2017b; *Liza ramada,* Rebaya *et al.*, 2017, *Sardinops sagax,* Izzo *et al.*, 2017). We report, below, the abstracts of the papers that apply otolith shape only and those that apply otolith shape in combination with other techniques are reported in the *Interdisciplinary Analysis* section.

Bacha *et al.* (2016) examined the geographic variability in otolith shape of round sardinella *Sardinella aurita* as a tool for stock discrimination. Fish were analysed from six sampling locations from Senegal to the Mediterranean coast of Morocco. A combination of otolith shape indices and elliptic Fourier descriptors was investigated by multivariate statistical procedures. Within the studied area, three distinct groups were identified with an overall correct classification of 78%. Group A: Nador (Alboran Sea), group B: Casablanca (northern Morocco) and group C: Senegalese–Mauritanian. The results of this study confirm the absence of an Atlantic Ocean–Mediterranean Sea transition for this species, the Gibraltar Strait acting as an efficient barrier for *S. aurita* population separation. Off north-west Africa, fish from northern Morocco form a single group which is clearly isolated from Senegalese–Mauritanian waters, confirming the existence of a distinct stock in this area. Among group C, some discontinuity exists and suggests the existence of a sedentary fraction of *S. aurita* in northern Mauritania (Arguin Bank). The results are discussed in relation to oceanographic features and physical barriers to dispersal and fish management strategy in the study area.

Bose *et al.* (2017) compared the morphology of sagittal otoliths of the plainfin midshipman fish *Porichthys notatus* between populations, sexes and male alternative reproductive phenotypes (known as 'type I males or guarders' and 'type II males or sneakers'). Sagitta size increased with *P. notatus* size and changes in shape were also detected with increasing body size. *Porichthys notatus sagittae* begin as simple rounded structures, but then elongate as they grow and take on a more triangular and complex shape with several prominent notches and indentations along the dorsal and caudal edges. Moreover, the *sagittae* of the two geographically and genetically distinct populations of *P. notatus* (northern and southern) differed in shape. *Porichthys notatus* from the north possessed taller *sagittae* with deeper caudal indentations compared to *P. notatus* from the south. Sagitta shape also differed between females and males of the conventional guarder tactic. Furthermore, guarder males had smaller *sagittae* for their body size than did sneaker males or females. These differences in sagittal otolith morphology are discussed in relation to ecological and life history differences between the sexes and male tactics of this species. This is the first study to investigate teleost otolith morphology from the perspective of alternative reproductive tactics.

Bostanci *et al.* (2016) studied the morphology, biometry, and shape indices of the left and right sagittal otoliths for sharpsnout seabream, *Diplodus puntazzo* species from Aegean Sea. The shape, *sulcus acusticus* shape, proximal and distal surfaces, anterior and posterior regions of left and right sagittal otoliths for a total of 52 *D. puntazzo* were analyzed. The morphometric measurements such as weight, length, width, area, and perimeter were recorded for each pair of sagittal otoliths of the sharpsnout seabream. The shape indices such as form factor, roundness, aspect ratio, circularity, rectangularity, and ellipticity were calculated for left and right sagittal otoliths of *D. puntazzo*. The otolith width and ellipticity were significantly different ($P < 0.05$) for left and right sagittal otolith measurements and shape indices, respectively in *D. puntazzo* inhabiting the Aegean Sea. Morphological characteristics of fish otoliths were highly variable in species and populations; there was limited information on the sagittal otolith morphology and shape indices. The present study provided sufficient information of the sharpsnout seabream left and right otolith morphologies, biometry, and shape indices in the Aegean Sea, that they may provide a useful tool for marine and freshwater species discrimination and identification in further investigations.

Harbitz (2016) studied parameter-sparse modification of Fourier methods to analyse the shape of closed contours with application to otolith outlines. Elliptical Fourier descriptors (EFDs) have been used extensively in shape analysis of closed contours and have a range of marine applications, such as automatic identification of fish species and discrimination between fish stocks based on EFDs of otolith contours. A recent method (the 'MIRR' method) transforms the two-dimensional contour to a one-dimensional function by mirroring (reflecting) the lower half of the contour around a vertical axis at the right end of the contour. MIRR then applies the fast Fourier transform (FFT) to the vertical contour points corresponding to equidistant coordinate values along the horizontal axis. MIRR has the advantage of reducing the number of Fourier coefficients to two coefficients per frequency component compared with four EFDs. However, both Fourier methods require several frequency components to reproduce a pure ellipse properly. This paper shows how the methods can be easily modified so that a virtually perfect reproduction of a pure ellipse is obtained with only one frequency component. In addition, real otolith examples for cod (*Gadus morhua*) and Greenland halibut (*Reinhardtius hippoglossoides*) are used to demonstrate that the modified methods give better approximations to the large-scale shape of the original contour with fewer coefficients than the traditional Fourier methods, with negligible additional computing time.

Ibanez *et al.* (2017) compared the discrimination of phenotypic stocks between otolith and scale shapes for *Mugil curema* specimens collected at five different locations in the Gulf of Mexico and two locations along the Pacific coast during two consecutive years. Geometric morphometric methods were used to determine the discrimination among locations using seven and 22 landmarks for scales and otoliths, respectively. The cross-validated discriminant analysis by location correctly classified 43.2 and 40.2% based on shape variables (Principal Components scores) for otoliths for all locations jointly, while for scales the classification percentages were 48.7 and 47.4% for the first and second years, respectively. Classification results improved when the discrimination analyses were carried out for pairs of locations, with 51.4 to 82.6% for otoliths and 72.7 to 97.1% for scales. The analysis was run for two consecutive years and the results for both years were best for the scales. Thus, fish scale shape offers a straightforward, non-destructive, accessible, quick and inexpensive method to trace fish phenotypic stocks.

Mapp *et al.* (2017) explored stock-separation of highly mobile Clupeids (sprat – *Sprattus sprattus* and herring – *Clupea harengus*) using otolith morphometrics. Analysis focused on three stock discrimination problems with the aim of reassigning individual otoliths to source populations using experiments undertaken using a machine learning environment known as WEKA (Waikato Environment for Knowledge Analysis). Six feature sets encoding combinations of size and shape together with nine learning algorithms were explored. To assess saliency of size/shape features half of the feature sets included size indices, the remainder encoded only shape. Otolith sample sets were partitioned by age so that the impact of age on classification accuracy could be assessed for each method. In total we performed 540 experiments, representing a comprehensive evaluation of otolith morphometrics and learning algorithms. Results show that for juveniles, methods encoding only shape performed well, but those that included size indices held more classification potential. However, as fish age, shape encoding methods were more robust than those including size information. This study suggests that methods of stock discrimination based on early incremental growth are likely to be effective, and that automated classification techniques will show little benefit in supplementing early growth information with shape indices derived from mature outlines.

Mille *et al.* (2016) investigated the potential correlation between diet and otolith shape in 5 wild marine fish species by addressing 4 complementary questions. First, is there a global relationship between diet and otolith shape? Second, which prey categories are involved in this relationship? Third, what are the respective contributions of food quantity and relative composition to diet–otolith shape co-variation? Fourth, is diet energetic composition related to otolith shape? For each species, they investigated how otolith shape varies with diet. These questions were tackled by describing diet in the analysis in 4 different ways, while also including individual-state variables to remove potential confounding effects. First, besides the strong effect of individual-state, a global relationship between diet and otolith shape was detected for 4 out of 5 fish species. Second, both main and secondary prey categories were related to variability in otolith shape, and otolith outline reconstructions revealed that both otolith global shape and its finer details co-varied with these prey categories. Third, the contribution of relative diet composition to diet–otolith shape co-variation was much higher than that of ingested food quantity. Fourth, the energetic composition of diet was related to otolith shape of only 1 species. These results suggest that diet in marine fish species may influence the quantity and composition of saccular endolymph proteins which play an important role in otolith biomineralization and their resulting 3D structure.

Rebaya *et al.* (2017) tested otolith shape discrimination of *Liza ramada* (Actinopterygii: Mugiliformes: Mugilidae) from marine and estuarine populations in Tunisia. The specimens of L. ramada were collected during three months (from March to May 2013) at two sampling sites: the marine (Cap Zebib sea resort) and the estuarine (Mellegue Dam) in Tunisia. They analysed sagittal otolith shape variation for 120 individuals (60 fish of each study site comprising 30 males and 30 females) for both sexes (males and females) and two sides (left and right otolith) for each specimen. Statistical- and discriminant function analysis of the sagittal otolith shape clearly demonstrated statistically significant differences from the two populations. These results were also confirmed by highly statistically significant difference between otolith shape (left and right) for both sexes. An asymmetry was detected when comparing otoliths of the same side (RR–LL) between different sampling sites. The shape variability of otolith between these two sampling sites is probably correlated with local environmental and ecological factors.

Wong *et al.* (2016) tested automated otolith image classification with multiple views. Combined multiple 2D views (proximal, anterior and ventral aspects) of the sagittal otolith are proposed here as a method to capture shape information for the classification. Classification performance of single view compared with combined 2D views show improved classification accuracy of the latter, for nine species of Sciaenidae. The effects of shape description methods (shape indices, Procrustes analysis and elliptical Fourier analysis) on classification performance were evaluated. Procrustes analysis and elliptical Fourier analysis perform better than shape indices when single view is considered, but all perform equally well with combined views. A generic content-based image retrieval (CBIR) system that ranks dissimilarity (Procrustes distance) of otolith images was built to search query images without the need for detailed information of side (left or right), aspect (proximal or distal) and direction (positive or negative) of the otolith. Methods for the development of this automated classification system are discussed.

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Otolith Chemistry (Contributors: Lisa Kerr and Dave Secor)

In the past year, otolith chemistry has been applied as a stock identification tool to discern stock structure of fish species around the world. Below is a summary of recent applications of otolith chemistry to fish stock identification of ICES species of interest, as well as an update on advances in the field.

Three papers utilized otolith chemistry to understand connectivity of fish species in an around marine protected areas. Lazartigues *et al.* (2016) applied otolith microchemistry to determine the source of capelin to a marine protected area in Canada (Saguenay-St. Lawrence Marine Park). Results indicated that the principal source of capelin to the marine park was the St. Lawrence estuary, outside the conservation area boundaries. Gibb *et al.* (2016) examined connectivity in the lesser sandeel, *Ammodytes marinus*, using otolith microchemistry within areas subjected to spatial management. Otoliths from juveniles were examined from four Scottish spawning areas predicted from modelled estimates of larval dispersal to differ in terms of larval retention rates and connectivity. Results revealed that one area (Firth of Forth) had a largely separate natal source. Regnier *et al.* 2017 analysed the otolith chemical signature the roundnose grenadier (*Coryphaenoides rupestris*) to evaluate the level of connectivity between the Rosemary Bank seamount, considered for inclusion in a network of MPAs, the Scottish west coast, and two adjacent locations. The elemental signatures of the fish from the seamount were distinguishable from the fish from the two other areas and authors concluded that once juveniles settled on the seamount they remain there for the rest of their lives.

Thomas *et al.* (2017) compared the elemental composition of otoliths versus endolymph. The authors applied size exclusion chromatography-inductively coupled plasma-mass spectrometry (SEC-ICP-MS) of endolymph to determine the binding interactions for a range of elements. The authors also applied solution ICP-MS to quantify element concentrations in paired otolith and endolymph samples and determined relative enrichment factors for each. The authors concluded that elements occurring only in the salt fraction are most likely to reflect changes in the physico-chemical environment experienced during life; elements occurring only in the proteinaceous fraction are more likely to reflect physiological rather than environmental events; and elements occurring in both the salt and proteinaceous fractions are likely to be informative about both endogenous and exogenous processes.

A special issue entitled *Frontiers in otolith chemistry: insights, advances and applications* was published in Journal of Fish Biology in 2017. The Special Issue grew out of a symposium held at the 2015 American Fisheries Society annual meeting in Portland, Oregon. The Special Issue consisted of nine manuscripts, including an overview of the special issue (Walther and Limburg 2017). Several of the contributions point out the necessity of validating basic assumptions about the chemical composition of otoliths as well as the selected analytical approach. In addition to otolith chemistry studies, a rapidly expanding area of interest is the use of alternative biological structures that may substitute as analogues for otoliths. These include scales, fin rays, vertebrae, scutes, and eye lenses. The papers are summarized below.

Limburg and Elfman (2017) highlighted the potential for heterogeneity in composition of sectioned otoliths in elements such as strontium, barium, manganese and selenium, a result relevant to analyzing otolith transects. Pracheil *et al.* (2017) investigated the composition of acipenserid otoliths and found that the crystal form was heterogenous with up to a third of the otoliths composed of calcite and the remainder vaterite, depending on the species. This study emphasized the need to carefully examine the underlying assumptions when interpreting otolith-derived migratory or environmental histories in fish. Jones *et al.* (2017) assessed alternative classification methods, including discriminant function analyses (linear and quadratic) and machine-algorithm methods. When parametric assumptions were met, the traditional parametric classification methods performed best, indicating that when data can be transformed to meet assumptions parametric classifiers should be used. This work highlights the need to carefully consider the statistical methods employed to classify unknown-origin individuals. Humston *et al.* (2017) coupling model-derived estimates of spatial heterogeneity in water ^{87}Sr:^{86}Sr ratios (Sr 'isoscapes') with field-collected water samples to assess model performance in a mainstem and tributary system. Water samples verified model isoscape predictions, and the study employed these markers to assess relative contributions of tributary spawning habitat and exchange among these two habitat types for a migratory centrarchid species.

In addition to otolith chemistry studies, a rapidly expanding area of interest is the use of alternative biological structures (e.g., scales, fin rays, vertebrae, scutes, and eye lenses). McMillan *et al.* (2017) discuss the assumptions and limitations of these approaches similar to the assessment of Elsdon *et al.* (2008) for otoliths. McMillan *et al.* (2017) highlighted the fact that elemental analyses in elasmobranchs is currently a young field, with few experimental validations of uptake and incorporation dynamics. As such, investigators must be cautious not to apply otolith-derived uptake dynamics to these structures and are advised to continue experimental assessments to allow confidence in interpreting patterns from wild-caught specimens.

Three papers in this Special Issue validated the utility of otolith analogues and employed them to infer migratory movements of wild-captured fishes. Bock *et al.* (2017) quantified relationships of elemental and isotope ratios between water and dentary material in paddlefish *Polyodon spathula* (Walbaum 1792), and found strong support for the use of these markers in reconstructing water chemistry history with these structures. Tzadik *et al.* (2017) investigated elemental and isotope ratio profiles across fin rays of a number of species and found strong concordance in patterns of some constituents between otoliths and fin rays from the same individuals, supporting the utility of fin rays as a non-lethal alternative to sampling otoliths. Phelps *et al.* (2017) employed elemental signatures in acipenserid fin rays sampled non-lethally to determine movement of individuals between reaches of large river systems. The use of non-lethal alternatives to otoliths is critical for these types of vulnerable species where mortality must be avoided. A review by Carlson *et al.* (2017) evaluated the application of otolith chemistry as a fisheries management tool, including its application to resolving questions of stock identification. The review demonstrated that otolith chemistry has diverse implications and applications for fisheries management worldwide.

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Parasites (Contributor: Ken Mackenzie)

In the past year, nine papers were published related to the use of parasites as biological tags in studies of the population structure of marine fish.

Three papers from the southwest Atlantic followed up on previous studies by Argentinian and Brazilian researchers who had identified different biogeographic regions in this area, each characterized by its own distinctive fauna of generalist parasites. Braicovich *et al.* (2016) referenced the use of parasites in fish stock assessment studies and assessed the role of different host traits (size, mass, age and their interactions with sex) as drivers of the abundance of long-lived parasites in this area. The results of generalised linear mixed models indicated fish length as a slightly better predictor than age or mass, leading the authors to recommend that comparisons of parasite abundance should be restricted to fish of similar length. The Brazilian flathead *Percophis brasiliensis* was the target host in both that study and another by Braicovich *et al.* (2017) which evaluated the utility of long-lived larval parasites (digeneans, cestodes, nematodes and acanthocephalans) as indicators of the previously identified zoogeographical regions off the coasts of Argentina, Uruguay and Brazil. Multivariate analyses showed a close fit between parasite assemblages and the existing zoogeographical classifications. The third paper from this region (Lanfranchi *et al.* 2016) focused on ecotonal regions – regions of convergence of different water masses – and analysed data on assemblages of long-lived larval parasites of silvery John Dory *Zenopsis conchifer* caught in the ecotonal region between two current systems. The results showed that parasite communities can be used as reliable indicators to define such regions, which are usually subjected to strong fishery pressures requiring the implementation of good management programmes.

Two papers examined parasites of fish in South African waters to address questions of stock identity. Nunkoo *et al.* (2016) investigated the community ecology of snoek *Thyrsites atun* in the Benguela system with respect to area, seasonality and life history stage of the host species. The homogeneity of the community structure of long-lived endoparasites suggested a single stock of snoek off South Africa. De Moor *et al.* (2017) followed up previous studies on the use of a “tetracotyle”-type metacercaria for stock identification of sardine *Sardinops sagax* off the west and south coasts of South Africa. New data enabled the authors to provide more precise estimates of annual movement and the extent of mixing of different age-groups of sardine between two different stocks.

In Mediterranean waters, Feki *et al.* (2016) used differences in the occurrence of 10 parasite taxa to identify three distinct areas within the distribution of juvenile and young adult horse mackerel *Trachurus trachurus* off the coast of Tunisia. In the North Atlantic, Klapper *et al.* (2017) investigated the spatial and temporal occurrence of the parasitic copepod *Sphyrion lumpi* on beaked redfish *Sebastes mentella* in the Irminger Sea. The authors found that abundance of *S. lumpi* remained constant during summer over the period 2001 to 2015, confirming its validity as a biomarker. Two stock units were identified, supporting continuation of the current management strategy.

Violante-González *et al.* (2016) surveyed the parasite faunas of green jack *Caranx caballus* from three locations off the Pacific coast of Mexico and evaluated their utility as biological tags. Of the 24 parasite taxa recorded, they identified 8 as being potentially useful tags. Multivariate discriminant analyses indicated the existence of three separate stocks with no evidence of migration between the three sampling locations.

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Interdisciplinary analysis (Contributors: All Participants)

Using interdisciplinary methods to investigate stock identity is a continuing trend in the field. Below we have summarized applications which involved multiple techniques applied to address questions of stock identity.

Boudinar *et al.* (2016) investigated genetic differentiation and species delimitation among nine *Atherina boyeri* populations from several marine and lagoon/brakish habitat sites in Algeria, Tunisia and France using three mitochondrial (control region, Cyt b and 16S) and one nuclear markers (2nd intron of S7). Five groups were found. Two of them perfectly corresponded to two species already recognized *Atherina presbyter* and *Atherina hepsetus*, both living in marine waters; and three additional, including *Atherina boyeri* (brackish and freshwater environments) and two independent groups of marine punctated and unpunctated individuals. Those findings are corroborated by the study of the otolith contour shape of 362 individuals of seven populations from different habitats using Fourier analysis. Samples from Ziama inlet, marine punctuated individuals and unpunctuated marine specimens from Annaba's Gulf formed three well separated groups. Specimens from Mellah and Mauguio lagoons formed another group. The last one includes individuals from Bizerte and Thau lagoons. The divergences between them strongly support the potential species within the *A. boyeri* species complex.

Callicó Fortunato *et al.* (2017a) characterized juvenile *Mugil cephalus* (flathead grey mullet) habitats in the Valencian community using otolith morphometry and microchemistry. Morphometric results showed, by an ANOVA with Bonferroni contrasts, that saccular otoliths from AV individuals had more edge complexity, hence a higher circularity index ($p < 0.001$), but that there was less otolith percentage occupied by the sulcus ($p < 0.001$). When analyzing the morphometric variables simultaneously, both sites differed significantly (Hotelling's T2 < 0.001). A paired t-test among sites of the microchemical variables showed that otoliths of AV presented higher values of Ba/Ca ratios and lower Sr/Ca ratios ($p < 0.001$). This coincides with water values obtained and could be associated with the low salinity observed in the lake. The opposite pattern was observed in SP, both for otolith and water samples, this being associated with the high-salinity waters of the area. Results obtained in the present research suggest, by the use of otolith morphometry and microchemistry that the nursery grounds of juvenile *M. cephalus* in the Valencian community could be differentiated. Even though habitats could be separated using otolith morphometry, only a few of the studied shape indices were important in area differentiation. Nevertheless, the use of both methodologies simultaneously could be robust habitat markers for this species.

Izzo *et al.* (2017) integrated genetic, morphological, otolith, growth, reproductive and fishery data collected over 60 years using a Stock Differentiation Index (SDI). The absence of strong separation (SDI[0.66) of most adjacent sub-groups supports the hypothesis that sardine (*Sardinops sagax*) in Australian waters is a meta-population, but with effective isolation of at least four stocks: south western coast (off Western Australia); Great Australian Bight and Spencer Gulf; Bass Strait and Port Phillip Bay (off Victoria and Tasmania); and eastern Australia. There is also evidence for sub-division of the stocks off Western Australia and the east coast. They examined age-related and inter-annual patterns of stock structure off South Australia and the east coast through integrated analysis of otolith chemistry and shape data. For the east coast, there were significant differences between northern and southern sub-groups for all three age cohorts examined. Fish were correctly classified to sampling region with a high degree of success ([80%), supporting the sub-division of the east coast stock suggested by the SDI. For South Australia, there were significant differences among two sub-groups for most cohorts examined across two sampling years. However, spatial discriminatory power was poor, with allocation success ranging from 48 to 64%. Results suggest that movements between the two South Australian sub-groups may vary among years, which is consistent with inconclusive SDI (0.5). Integrating historical data using a SDI is suitable for identifying fishery management units. Integrated analysis of otoliths from archival collections is useful for examining temporal variability in stock structure, which is also important for fisheries management. Our findings are relevant to fisheries where sustainability risks are increased by management arrangements based on assumptions that stock structure is absent or stable.

Callicó Fortunato *et al.* (2017b) investigated the mullet *Mugil liza* that lives southernmost in the western Atlantic Ocean. Knowledge about migration, movements and identification of stocks of this important fishery resource is scarce. Thus, they aimed to study movement patterns and to identify the presence of different fish stocks in the southwestern region of the Atlantic Ocean, using cumulative otolith shape morphometric and microchemical analyses of sagittae otoliths. Specimens (n = 99) were obtained in four coastal areas: Paranaguá Bay in Brazil, Samborombón Bay, Mar Chiquita Coastal Lagoon, and San Blas Bay in Argentina. Otolith shape indices (circularity, rectangularity, aspect ratio, percentage occupied by sulcus, ellipticity and form factor) were used for stock identification analysis; and otolith microchemistry using LA-ICP-MS (Sr/Ca and Ba/Ca ratios chronological variation) was used for both the analysis of movement behaviors and the identification of fish stocks (otolith edge ratios). Morphometrical indices did not reveal a clear separation among areas. San Blas bay individuals presented otoliths tending to be longer than wider, with a more elliptic shape than the otoliths from other studied areas; also, this area did not share individuals with the most northern one, Paranaguá Bay in Brazil. The analysis of microchemical lifetime profiles revealed three types of behavior pattern: Type I: most frequent use of estuarine environments; Type II: a fluctuating behavior between estuarine and sea/high salinity waters; Type III: most frequent use of sea/high salinity habitats. Otolith edge analysis did not reveal differences among Sr/Ca and Ba/Ca ratios for the different areas. Thus, it cannot be assured that there is more than one stock in the studied region. *Mugil liza* revealed different environmental migratory behaviors in the Southwestern Atlantic Ocean showing a facultative use of estuarine waters; hence, the species appears to be mostly coastal with the use of low estuaries, as seen also by the Sr/Ca otolith cores ratios; differing from the general mugilid behavior previously described.

Lowerre-Barbieri *et al.* (2016) used a combination of acoustic tagging data with aerial surveys and capture statistics to study migration patterns and stock structure in juvenile and adult red drum off Florida, USA. They documented strong spatio-temporal patterns in spawning site selection with population-specific spawning site fidelity and natal homing.

Taillebois *et al.* (2017) applied parasite tagging, together with genetics and otolith chemistry, used in a study of the stock structure of the sciaenid fish *Protonibea diacanthus* in coastal waters of northern Australia (Taillebois *et al.* 2017). All three methods suggested strong spatial variation between sampling locations, indicating the existence of a number of locally discrete populations with restricted exchange between them.

Duarte *et al.* (2017) investigated the composition of Caranx spp. catch from Rio de Janeiro State, Brazil with classical taxonomic, multivariate morphometry, and allozyme molecular markers to determine that at least three species were present. However, using Bayesian methods, it was determined that there is evidence to suggest that four species may be present, whereas only two species are recorded in commercial fisheries landings reports, complicating the management of the fishery.

Parsons *et al.* (2016) tested the use of morphological and meristic characteristics for stock identification of snapper off New Zealand with the combined use of otolith chemistry and tag-recapture data from taggings with external dart tags. While no morphological differences were observed between stock components, otolith chemistry and tag-recapture information showed strong geographical complexity in population structure, highlighting the need to revisit spatial stock management units.

Ulrich *et al.* (2017) evaluated stock management units of plaice in the North Sea- Skagerrak – Kattegat area using the combined information from a century of T-bar tagging data coupled with genetics, otolith growth and hydrogeographical drift modelling. Considerably connectivity in populations between Skagerrak and the North Sea resulted in these two management units being merged into a single one.

Pérez-Quiñónez *et al.* (2017) used a combined method of geometric morphometrics and genetic analysis to identify species within the *Opisthonema* complex of Pacific threadfin herring in the eastern Mexican Pacific. These species are harvested together in mixed aggregations and proper determination of catch share is important to future management.

Ozerov *et al.* (2016) combined genetic markers with the traditionally utilized gill-raker count to improve mixed-stock analysis in Baltic whitefish (***Coregonus lavaretus s.l.***) to identify sea- and river-spawning ecotypes.

Vieira *et al.* (2016) analyzed specimens of forkbeard *Phycis phycis* from the Northeast Atlantic Ocean (Azores, Madeira, and mainland Portugal) for geometric morphometric analysis and genetic differences. Additional genetic samples from Spain, Italy, and Croatia were analyzed to effectively cover the entire range of the forkbeard. Although the morphology among the three locations was found to be highly variable and hypothesized to be due to environmental factors, genetic differentiation overall was low. The authors conclude that this is possibly indicative of recent population expansions to the Northeast Atlantic from the Mediterranean Sea during the Last Glacial Maximum.

Stern *et al.* (2017) conducted a global study to determine the differentiation among the five putative sardine species within the subgenus *Sardinella*, including morphological and genetic analyses. Morphometric analysis found only two morphospecies and the three species-delimination analyses failed discriminated the five putative species, with the authors suggesting a single global cosmopolitan species *Sardinella aurita* with ecophenotypic variations. This study calls for the merging of phylogenetic and population genetic approaches to test the feasibility of taxonomic theory relating to this species. This in turn underscores the importance validating basic species identification with the use of typical stock identification methods.

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Published Theme Set Update (Contributor: Lisa Kerr)

In last year's SIMWG report, we referenced the plan to publish a theme set entitled: *Beyond Ocean Connectivity* in the ICES *Journal of Marine Science* that followed up a session by the same name at ICES ASC 2015 (Conveners: Manuel Hidalgo, Lisa Kerr, and Claire Paris). The theme set was published in July 2017 (volume 74, Issue 6) and contained nine papers that were grouped into four topic areas: methodological advances, population dynamics and assessment implications of connectivity, spatial and management implications, and connectivity in ecosystem processes.

Methodological advances - Kaplan *et al.* (2017) developed probabilistic statistical methods for estimating uncertainty of a broad range of larval mark-recapture connectivity estimators, including otolith microchemistry and genetic parentage analysis. Additionally, the use of larval mark-recapture experiments to estimate marine connectivity was explored in two studies, one using artificial chemical tagging of larval otoliths (Secor *et al.*, 2017), the other applying genetic approaches to assign recruits to source populations (Christie *et al.*, 2017). In other methodological advances, Christie *et al.* (2017) applied a forward-time agent-based model of genetic information to Kellet's whelk (*Kelletia kelletii*) in southern California, incorporating key life history and physical oceanographic information, to assess the advantages and disadvantages of two commonly used methods to assign individuals back to their natal origin. Monroy *et al.* (2017) presented an assessment of Lagrangian Flow Networks, a numerical modelling technique to estimate larval connectivity.

Population dynamics and assessment implications of connectivity - Kerr *et al.* (2017) presented a review of approaches applied to resolve mismatches between the scale of biological populations and spatially defined stock units. The review included case studies that applied the following approaches: (i) status quo management, (ii) "weakest link" management, (iii) spatial and temporal closures, (iv) stock composition analysis, and (v) alteration of stock boundaries.

Spatial and management implications - Gallego *et al.* (2017) investigated the connectivity of epi-benthic species as a part of the Scottish nature conservation MPA designation process. Nicolle *et al.* (2017), examined great scallop (*Pecten maximus*) dispersal pathways and the patterns of connectivity between fishing grounds in the English Channel. Zemeckis *et al.* (2017) investigated the seasonal movemrn Gulf of Maine.

Connectivity in ecosystem processes - Lough *et al.* (2017), illuents and connectivity of the spring spawning component of Atlantic cod in the westestrated how prey selection by juvenile pelagic cod for zooplankton of different size (*Centropages spp.* vs. *Pseudocalanus spp.*) under different climatic regimes can affect growth and survival rates.

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