



# Fish and Sclerochronology Research in the Mediterranean: Challenges and Opportunities for Reconstructing Environmental Changes

Sanja Matić-Skoko<sup>1\*</sup>, Melita Peharda<sup>2</sup>, Dario Vrdoljak<sup>1</sup>, Hana Uvanović<sup>2</sup> and Krešimir Markulin<sup>3</sup>

<sup>1</sup> Laboratory of Ichthyology and Coastal Fisheries, Institute of Oceanography and Fisheries, Split, Croatia, <sup>2</sup> Laboratory of Fisheries Science and Management of Pelagic and Demersal Resources, Institute of Oceanography and Fisheries, Split, Croatia, <sup>3</sup> Laboratory of Chemical Oceanography and Sedimentology of the Sea, Institute of Oceanography and Fisheries, Split, Croatia

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### \*Correspondence:

Sanja Matić-Skoko  
sanja@izor.hr

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Over the past two decades, the field of sclerochronology has been rapidly developing, with scientists devoting significant efforts to studying the physical and chemical variations in hard tissues of aquatic organisms. Most of this research has been limited to certain taxa and geographic areas. Although growth increments in fish otoliths are used for sclerochronology purposes, relatively little has been done in the Mediterranean Sea. According to the literature, the chemical composition of otoliths from Mediterranean fish species has primarily been used for analyzing migration patterns, habitat use, and population structure of commercially important fish species. To the best of our knowledge, there are no studies on fish growth chronology construction conducted in the Mediterranean Sea. In order to identify the opportunities for sclerochronology research on fish from the Mediterranean, we used FishBase to identify potential candidate species with a sufficiently long lifespan and clearly defined growth increments for growth chronology construction and otolith chemistry research. We also present the challenges and limitations for sclerochronology research, including: (i) very few fish species in the Mediterranean Sea have a longevity of several decades; (ii) issues associated with reliable age determination for certain long-lived fish species; (iii) a general lack of understanding and effort to construct and manage otolith collections; and (iv) limitations imposed by the availability of funding, expertise, and instrumentation. Despite these challenges, fish sclerochronology research has strong potential in the Mediterranean and adjacent seas. Recent studies in the Adriatic Sea have resulted in the construction of bivalve chronologies and the geochemical analysis of shells, providing important time-series data for comparative analysis and a multispecies approach. Furthermore, studies conducted in other parts of the world have demonstrated great potential for the use of fish otoliths in monitoring environmental variability and the effects of pollutants and disturbance.

**Keywords:** otolith, sclerochronology, growth increments, geochemical fingerprints, stable isotopes, longevity, Mediterranean Sea

## INTRODUCTION

Hard structures of aquatic organisms, including mollusk shells, fish otoliths, corals, and coralline algae, are deposited continuously during the life of the organism, and thereby contain environmental information collected over the organism's life cycle (e.g., Hudson et al., 1976; Jones, 1983; Black et al., 2008). The field of sclerochronology utilizes these data archives by investigating their morphological (i.e., increment width) and geochemical composition to deduce organismal life history traits as well as to reconstruct records of environmental and climatic change through space and time (Oschmann, 2009). Although many sclerochronology studies have been conducted on sedentary organisms, primarily the bivalve *Arctica islandica* (e.g., Schöne, 2013; Marali et al., 2017; Reynolds et al., 2018), fish also present a very interesting target taxon for sclerochronology research (e.g., Panfili et al., 2002; Black et al., 2005; Grønkvjaer et al., 2013). The objective of this review was to focus primarily on papers that analyzed otoliths in relation to environmental and climatic changes. There are numerous studies in the review that analyzed fish growth and age from growth increment structures in the otoliths (e.g., Gutiérrez and Morales-Nin, 1986; Morales-Nin and Moranta, 1997; Reñones et al., 2007), but they did not directly relate them to the environmental conditions and a detailed review of such studies is beyond the scope of a present paper.

One of the challenges in evaluating the status of sclerochronology research lies within the fact that this term is not always used in publications addressing the morphological and/or geochemical properties of hard structures in aquatic organisms (Gillikin et al., 2019). This is especially the case for research conducted on fish, despite the publication of the very comprehensive *Manual of Fish Sclerochronology* (Panfili et al., 2002).

Fish possess several hard structures interesting for sclerochronology analysis, including scales, the skeleton, and otoliths (e.g., Chilton and Beamish, 1982; Panfili et al., 2002). Of these, otoliths—calcium carbonate structures located in the inner ear of the fish—are considered the most reliable, as they are metabolically inert, hindering re-absorption (Campana and Neilson, 1985), unlike other structures, such as scales (Simkiss, 1974). Otoliths contain periodically deposited growth increments, from daily to annual, and can thereby provide high temporal resolution data (e.g., Campana, 1999; Morales-Nin, 2000; Black et al., 2008; Elsdon et al., 2008). As fish can attain a maximal life span of several decades, otolith analysis can provide an important window into the past (e.g., Campana, 1999; Black et al., 2008).

Chemical research on otoliths includes analysis of elemental and/or isotopic composition. In 1999, Campana published a review paper on the chemistry and composition of otoliths, presenting in detail the state of the art on this subject at that time and the applications and assumptions of this type of research. The applications of otolith chemistry for describing movements and life-history parameters of fish were comprehensively presented by Elsdon et al. (2008). Numerous publications followed, clearly demonstrating the potential for otolith chemistry as a natural

tag of fish stocks (e.g., Trueman et al., 2012; Darnaude and Hunter, 2018; Izzo et al., 2018; Wright et al., 2018). Although most studies focus on stock identification and migration history, the elemental composition of otoliths can also be applied for identifying bioavailable contaminants and establishing long-term trends (e.g., Søndergaard et al., 2015; Andronis et al., 2017; Mounicou et al., 2019). Furthermore, as oxygen isotopes ( $\delta^{18}\text{O}$ ) are considered a proxy of water temperatures, analysis of otolith isotopic composition can enable reconstruction of environmental conditions (e.g., West et al., 2012; Willmes et al., 2019).

Use of otolith growth increments to construct fish growth chronology and establishing the relationship with environmental conditions have received increasing attention over the past decade. The methodology for this research has been derived from dendrochronology—the study of growth rings in trees (Black et al., 2005). Primary target organisms are long-living fish species, such as yelloweye rockfish (*Sebastes ruberrimus*, >70 years; Black et al., 2008), and northern rockfish (*Sebastes polyspinis*, ~40 years; Matta et al., 2018). However, development of statistical methods and sample archives have also enabled growth chronology construction for shorter living species. For example, Tanner et al. (2019) constructed half a century chronology for a small, relatively short-lived (<16 years) pelagic fish (Atlantic horse mackerel, *Trachurus trachurus*).

The main objective of this paper is to present an overview of the sclerochronology related research in the Mediterranean Sea and to present its opportunities and challenges.

## OVERVIEW OF PREVIOUS SCLEROCHRONOLOGY RELATED RESEARCH IN THE MEDITERRANEAN

In order to identify relevant publications on fish sclerochronology research in the Mediterranean Sea, we conducted a literature search through the Web of Science database. The keywords “Mediterranean” and “otolith” were used in combination with words “isotope,” “element,” “microchemistry,” “chemistry,” and “chronology.” All publications obtained through this search were read in detail, and only those relating to otolith analysis were included (Table 1 and Supplementary Table S1). Other structures, such as scales and vertebrae, were not considered for the purposes of this review.

Chemical analysis of otoliths has been conducted on over 41 fish species from the Mediterranean, and the most studied species are from the family Sparidae (Table 1). Published studies include data for the entire otolith (e.g., Iacumin et al., 1992; Gillanders et al., 2001; Marini et al., 2006; Arechavala-Lopez et al., 2016), data for a specific area of the otolith (e.g., Tanner et al., 2012; Mirasole et al., 2017; Rooker et al., 2019), and time series data (e.g., Correia et al., 2012; Mercier et al., 2012; Bouchoucha et al., 2018). In most reports, only a single species was analyzed, while Papadopoulou and Moraitopoulou-Kassimati (1977), Iacumin et al. (1992), Marini et al. (2006), Swan et al. (2006), Khemiri et al. (2014), Arechavala-Lopez et al. (2016), Mirasole et al. (2017), Bouchoucha et al. (2018), and Demirci et al. (2018) presented data for 2 to 24 different species.

**TABLE 1** | List of Mediterranean fish species targeted in the otolith microchemistry research.

Family	Species	References
Anguillidae	<i>Anguilla anguilla</i>	Panfili et al., 2012; Capoccioni et al., 2014
Carangidae	<i>Trachurus mediterraneus</i>	Turan, 2006
	<i>Trachurus trachurus</i>	Iacumin et al., 1992
Centracanthidae	<i>Spicara maena</i>	Iacumin et al., 1992
Champsodontidae	<i>Chamsodon nudivittis</i>	Demirci et al., 2018
Clupeidae	<i>Sardina pilchardus</i>	Iacumin et al., 1992; Khemiri et al., 2014
Congridae	<i>Conger conger</i>	Papadopoulou and Moraitopoulou-Kassimati, 1977; Correia et al., 2011, 2012
Engraulidae	<i>Engraulis encrasicolus</i>	Iacumin et al., 1992; Marini et al., 2006; Guidetti et al., 2013; Khemiri et al., 2014
Gadidae	<i>Merlangus merlangus</i>	Papadopoulou and Moraitopoulou-Kassimati, 1977; Iacumin et al., 1992
	<i>Trisopterus minutus</i>	Iacumin et al., 1992
Gobiidae	<i>Gobius bucchichi</i>	Mirasole et al., 2017
	<i>Gobius niger</i>	Papadopoulou and Moraitopoulou-Kassimati, 1977
Labridae	<i>Coris julis</i>	Mirasole et al., 2017
	<i>Symphodus ocellatus</i>	Mirasole et al., 2017
Macrouridae	<i>Nezumia aequalis</i>	Swan et al., 2003, 2006
Moronidae	<i>Dicentrarchus labrax</i>	Iacumin et al., 1992; Traina et al., 2015; Arechavala-Lopez et al., 2016
Merlucciidae	<i>Merluccius merluccius</i>	Iacumin et al., 1992; Morales-Nin et al., 2005a; Swan et al., 2006; Tomas et al., 2006; Hidalgo et al., 2008; Tanner et al., 2012; Chang et al., 2012; Morales-Nin et al., 2014; Tanner et al., 2014; Vitale et al., 2016
Mugilidae	<i>Mugil cephalus</i>	Iacumin et al., 1992; Fortunato et al., 2017a,b
Mullidae	<i>Mullus barbatus</i>	Marini et al., 2006
Nemipteridae	<i>Nemipterus randalli</i>	Demirci et al., 2018
Pomacentridae	<i>Chromis chromis</i>	Mirasole et al., 2017
Sciaenidae	<i>Sciaena umbra</i>	Iacumin et al., 1992; Marini et al., 2006
	<i>Umbrina cirrhosa</i>	Marini et al., 2006
Scombridae	<i>Scomber japonicus</i>	Papadopoulou et al., 1978, 1980
	<i>Thunnus alalunga</i>	Davies et al., 2011
	<i>Thunnus thynnus</i>	Secor and Zdanowicz, 1998; Rooker et al., 2002, 2003, 2008a,b, 2014, 2019; Secor et al., 2002; Fraile et al., 2016
Sebastidae	<i>Helicolenus dactylopterus</i>	Swan et al., 2006
Serranidae	<i>Serranus scriba</i>	Iacumin et al., 1992
Sparidae	<i>Boops boops</i>	Iacumin et al., 1992
	<i>Diplodus annularis</i>	Papadopoulou and Moraitopoulou-Kassimati, 1977; Iacumin et al., 1992; Catalán et al., 2018
	<i>Diplodus puntazzo</i>	Iacumin et al., 1992

(Continued)

**TABLE 1** | Continued

Family	Species	References
	<i>Diplodus sargus</i>	Iacumin et al., 1992; Di Franco et al., 2011, 2012, 2015b; Bouchoucha et al., 2018
	<i>Diplodus vulgaris</i>	Iacumin et al., 1992; Gillanders et al., 2001; Di Franco et al., 2014, 2015a; Mirasole et al., 2017; Bouchoucha et al., 2018
	<i>Dentex dentex</i>	Iacumin et al., 1992
	<i>Lithognathus mormyrus</i>	Iacumin et al., 1992
	<i>Oblada melanura</i>	Iacumin et al., 1992; Calò et al., 2016
	<i>Pagellus erythrinus</i>	Iacumin et al., 1992; Marini et al., 2006
	<i>Sarpa salpa</i>	Iacumin et al., 1992; Mirasole et al., 2017
	<i>Sparus aurata</i>	Iacumin et al., 1992; Mercier et al., 2012; Tournois et al., 2013; Arechavala-Lopez et al., 2016; Niklitschek and Darnaude, 2016
	<i>Spondyliosoma cantharus</i>	Iacumin et al., 1992
Soleidae	<i>Solea solea</i>	Dierking et al., 2012; Morat et al., 2012, 2013, 2014a,b

Species names listed as valid species according to World Register of Marine Species. More detail on these studies is available in **Supplementary Table S1**.

Recently, Chang and Geffen (2013) summarized taxonomic and geographic influences on fish otolith microchemistry based on a number-published paper worldwide and all those related to the Mediterranean are also included in this study.

In total, 12 species from the family Sparidae are listed in **Table 1**, and most were addressed only by Iacumin et al. (1992). This study analyzed the oxygen and carbon isotope composition of aragonite in fish otoliths with regard to their possible suitability in paleoenvironmental and paleobiological work. This was the first attempt to apply stable isotope analysis to fish otoliths from the Mediterranean Sea. The mostly analyzed species are those from *Diplodus* genus, particularly *D. sargus*, and *D. puntazzo*, regarding possible environmental interpretation of otolith fingerprints related to spatial patterns of population connectivity and dispersal of marine fishes (Di Franco et al., 2011, 2015b), dispersal scales of fish at various life history stages, which is critical for successful design of networks of marine protected areas (Di Franco et al., 2012, 2015a), within-otolith variability enabling its usage as a marker for fish exposure to stressful conditions (Di Franco et al., 2014).

The majority of research on otolith chemical composition has been conducted on commercially important species. Atlantic Bluefin tuna (*Thunnus thynnus*) was the target of several studies on the element (Secor and Zdanowicz, 1998; Rooker et al., 2003) and isotope composition (Secor et al., 2002; Rooker et al., 2008a,b, 2014, 2019), aiming to reconstruct movement and population exchange. Fraile et al. (2016) observed the depletion in  $\delta^{13}\text{C}$  in *T. thynnus* otoliths over time, associating this with the oceanic uptake of anthropogenically derived  $\text{CO}_2$  from the Mediterranean Sea over the past two decades. These studies

primarily focused on material deposited in the otolith core or during the first year of life.

The European hake (*Mercuccius merluccius*) is also an important target species for the analysis of otolith element and isotope composition, and different methodologies have been applied. Morales-Nin et al. (2005a) studied elements in different parts of the otoliths using laser ablation–spot analysis, while in a study from 2014, the same authors used the line scan approach. Laser ablation, as opposed to otolith dissolution that is applied in the analysis of the whole otolith, enables the collection of more data points that can be placed in time (e.g., Elsdon et al., 2008). Tomas et al. (2006) studied composition of the opaque and translucent bands with wavelength dispersive spectrometry (WDS) revealing that annual marks (translucent) were significantly richer in Sr and Ca and significantly poorer in Na than opaque bands. Swan et al. (2006) applied two methods—solution-based inductively coupled plasma mass spectrometry of the whole otolith and laser ablation analysis of the otolith nucleus on hake and bluemouth (*Helicolenus dactylopterus*). Chang et al. (2012) used hake otoliths to test different widths of ablation lines and evaluate the temporal resolution of data. Hidalgo et al. (2008) and Tanner et al. (2012) applied analysis of  $\delta^{18}\text{O}$  and  $\delta^{13}\text{C}$  to certain sections of the otolith, specifically the core and the edge zone, in determining hake movement and ecology. It is interesting that Tanner et al. (2012) combined stable isotope analysis with analysis of otolith element composition to obtain more comprehensive data. These studies on hake analyzed the migration, population structure, and ecology of this commercially important species. In recent papers, Tanner et al. (2014) accompanied genotype with otolith data to increase the classification accuracy of individuals to their potential natal origins, while Vitale et al. (2016) estimated longevity of 25 years of female hake by applying bomb radiocarbon dating.

Element and isotope composition of otoliths of the common sole (*Solea solea*) have been analyzed in a series of studies conducted in the Gulf of Lions, in the northwest Mediterranean (Dierking et al., 2012; Morat et al., 2012, 2013, 2014a,b). These studies analyzed dispersion between populations and the use of different habitats.

The chemical composition of otoliths as a proxy of environmental conditions has been analyzed in only a few studies in the Mediterranean Sea, including Traina et al. (2015). They investigated the metal content of European sea bass (*Dicentrarchus labrax*) otoliths from two fish aquaculture sites. Their results indicated variations in the concentrations of certain metals between locations that were likely due to industrial effluents.

To the best of our knowledge, there is no research in the Mediterranean Sea related to fish growth chronologies constructed from growth increment analysis. The literature search conducted through the Web of Science returned just one publication for the keyword combination “Mediterranean” and “fish” and “sclerochronology”: a report by Prendergast and Schöne (2017) as the preface to the *Special Issue on Sclerochronology* containing research from different parts of the world including the Mediterranean, but not specifically on fish in the Mediterranean.

## OPPORTUNITIES FOR FISH GROWTH CHRONOLOGY CONSTRUCTION

Over the past two decades, techniques developed in dendrochronology research have been applied for the construction of fish growth chronologies (Black et al., 2005, 2008). They clearly demonstrated the potential for obtaining long term data from growth patterns in otoliths and for identifying environmental drivers (Morrongiello et al., 2012; Rountrey et al., 2014). In a recent review, Black et al. (2019) presented a global list of fish species that have been the subject of sclerochronology studies that included growth chronology construction and applied cross-dating techniques. Their list includes 21 species, none of which were from the Mediterranean Sea. The most studied on the list are cold-water species, such as kelp greenling (*Hexagrammos decagrammus*) and black rockfish (*Sebastes melanops*) and other species from the genus *Sebastes* (*S. alutus*, *S. aurora*, *S. diploproa*, and *S. ruberrimus*). Other species were from the families: Girellidae, Labridae, Lethrinidae, Lutjanidae, Platycephalidae, Pleuronectidae, Polyprionidae, Sciaenidae, and Scombridae. All of these are long-lived, non-migratory, nearshore residents with generalist diets that can be caught easily throughout a wide geographic range (Whitfield and Elliott, 2002).

Identifying target fish species with a sufficiently long life span and clearly defined growth increments is a prerequisite for statistically robust chronology construction. Unlike bivalves and trees that can live for several centuries (e.g., *A. islandica*, 507 years; Butler et al., 2013) or even millennia (e.g., *Pinus longaeva*, 4,900 years; Currey, 1965), fish have a shorter lifespan and present a greater challenge for constructing statistically robust chronologies. Another prerequisite for chronology construction is the availability of samples, which needs to take the conservation status of species into account. Although it is scientifically interesting to obtain data from endangered species, sampling such species should be clearly justified and ethically sound. Replication is essential for proper cross-dating that can yield annually resolved chronologies sensitive to environmental stressors (Hudson et al., 1976).

In order to identify possible candidates for fish growth chronology research in the Mediterranean Sea, we conducted a search of the FishBase database. This is a global, scientifically guided, biodiversity information system on fishes that provides a wide range of information (taxonomy, biology, trophic ecology, and life history) on all species currently known in the world, as well as historical data reaching back 250 years<sup>1</sup>. According to this database, a total of 755 fish species from 174 families inhabit the Mediterranean Sea. We made several reductions to obtain a reasonable pool of potentially interesting target species. Since the database provides data for maximal recorded total length (TL), maximal reported age, trophic level, and habitat p'nces (demersal, pelagic-neritic, benthopelagic, bathypelagic, bathydemersal, pelagic-oceanic, and reef-associated) and status (endemic, introduced, and native or questionable), we first removed all short-lived (<2 years) and

<sup>1</sup>fishbase.org



small fishes (TL < 30 cm). This resulted in the removal of species belonging to the families Apogonidae, Atherinidae, Blenniidae, Bregmacerotidae, Callionymidae, and Carapidae. Given their conservation status, Chondrichthyes and the primitive fishes (Myxinidae, Petromyzontidae, Chimaeridae, and Halosauridae) were also excluded. Furthermore, rare or poorly investigated species without any commercial interest or benthopelagic, bathypelagic, and bathydemersal fishes for which no age-related data were provided in FishBase were also excluded.

Finally, a pool of 263 fish species inhabiting Mediterranean Sea was obtained and used in the analysis. The estimated or determined maximum age of 31 fish species from 20 families was over 30 years. However, it is important to note that age was determined by age reading methods on specimens from the Mediterranean Sea for only 12 species (**Table 2**), while the age of other species was estimated based on growth equation parameters available for that specific species or closely related species from the same family, or age was reported for an area other than the Mediterranean Sea.

From these reports, the longest-living species in the Mediterranean Sea is the wreckfish, *Polyprion americanus* (see Peres and Haimovici, 2004). However, its maximum age of 76 years was reported for specimens from the continental shelf and slope off southern Brazil. Thus, its availability for sclerochronology studies in the Mediterranean is questionable, particularly since this species has been listed as Critically Endangered (CR) on the IUCN Red List (IUCN, 2017). The same is true for the red bream *Beryx decadactylus*, since the reported maximum age of 61 years was for specimens collected off the southeastern coast of the United States. To the best of our knowledge, there are no relevant data for the maximum age of either species in the Mediterranean.

The dusky grouper, *Epinephelus marginatus*, lives throughout the Mediterranean Sea and its maximum age of 60 years was reported for specimens from the Balearic Islands (Reñones et al., 2010). Most groupers are solitary, resident fishes. The Mediterranean is the upper limit of their northward distribution, and their growth in the Mediterranean is significantly slower than for groupers in tropical waters (Gracia\_López and Castelló-Orway, 2003). Site specificity, a relatively slow growth rate (some species may not be mature until the age of 8 to 10 years) and spawning strategy (synchronic or protogynous hermaphrodites; Sadovy and Shapiro, 1987; Heemstra and Randall, 1993) make them particularly vulnerable (CITES/UNEP-WCMC, 2017). Although the long-life span and resident behavior makes *E. marginatus* an interesting candidate for construction of growth chronologies, its low abundance and protected status throughout the Mediterranean requires a strategic approach to sample collection extending over time, rather than single on-site sampling action.

Three families listed in **Table 2**—Sebastidae, Lutjanidae, and Sciaenidae—were identified earlier within the list of globally important fish taxa for sclerochronology research (Black et al., 2019). However, just two species from the Sebastidae family are listed in the Mediterranean Sea, and only *H. dactylopterus* can attain an age of over 40 years. Certain caution is needed, as this data was reported for individuals caught in the Northeast Atlantic and not in the Mediterranean Sea. According to the

available data, *H. dactylopterus* grows faster and lives longer in the Northeast Atlantic than in the Mediterranean (Ragonese, 1989; Allain and Lorange, 2000; D'Onghia et al., 2004; Consoli et al., 2010). The maximum age reported for *H. dactylopterus* from the Mediterranean is 21 years (Consoli et al., 2010), questioning the availability of Mediterranean samples for growth analysis for this species.

Two species of Sciaenidae family are listed in **Table 2**. The maximum reported age for the meagre, *Argyrosomus regius*, in the Gulf of Cádiz (SW Iberian Peninsula) is 42 years (González-Quirós et al., 2011), while the brown meagre, *Sciaena umbra*, reached 31 years in the Gulf of Tunis (Chater et al., 2018). The dense calcium carbonate deposition of the large and very thick otoliths in Sciaenids reduces light transmission, making it almost impossible to distinguish hyaline and opaque zones (Arneri et al., 1998; Chater et al., 2018). According to Arneri et al. (1998), growth increments in otoliths of these taxa are more readable in cross-sections. Both species are commercially important and there is the potential for collection of representative otolith samples. However, further development of otolith reading techniques is needed to facilitate identification of growth increment boundaries and enable statistically robust chronology construction.

Two non-native species, yellowbar angelfish (*Pomacanthus maculosus*) and mangrove red snapper (*Lutjanus argentimaculatus*), have a lifespan of over 30 years (Grandcourt et al., 2004; Fry et al., 2006) and are interesting candidates for growth chronology construction. Both species entered the Mediterranean via the Suez channel and in recent years have established their populations in the eastern Mediterranean, along the coasts of Israel and Lebanon (Bariche, 2010; Sonin et al., 2019). The maximum reported age for *P. maculosus* is for specimens from the southern Arabian Gulf, while for *L. argentimaculatus* the maximum age data is reported for its native range—Papua New Guinea. These two species belong to long-living families (Grandcourt et al., 2004; Piddocke et al., 2015), and although determination of otolith growth patterns present certain challenges for the oldest specimens (Rezende and Ferreira, 2004; Steward et al., 2009), in the context of climate change they are interesting taxa for growth chronology research.

The remaining species listed in **Table 2** belong to the families Sparidae and Moronidae. There are total of 31 sparid species in the Mediterranean Sea, which are known to be slow-growing and long-lived (Hanel and Tsigenopoulos, 2011) and susceptible to over-exploitation due to their commercial importance (Comeros-Raynal et al., 2016). Sparid fishes generally have relatively large and easily readable sagittal otoliths, and despite the wealth of literature denouncing the use of whole, unsectioned otoliths in growth studies on sparid fishes (see Winkler et al., 2019), age determination using whole otoliths is still common. According to the information available in Fish Base, the maximum age reported for sparids in the Mediterranean ranges from 5 to 36 years (**Table 2**). Species with the greatest potential are common dentex *Dentex dentex* and zebra seabream *Diplodus cervinus*. Due to its commercial importance, wide distribution, clear growth patterns in otoliths, and lifespan of over 20 years (Kraljević et al., 1998), the gilt head seabream *Sparus aurata* is also an interesting candidate for sclerochronology research. From the Moronidae family, sea

**TABLE 2** | The list of long-lived fish species in the Mediterranean Sea according to FishBase. Data sorted by descending maximum reported age.

Family	Species	Length (cm)	Maximum age (year)	Trophic level	Habitat	References
Serranidae	<i>Epinephelus marginatus</i>	150.0	60	4.1	Reef-associated	Reñones et al., 2010
Carangidae	<i>Pseudocaranx dentex</i>	122.0	49	3.9	Reef-associated	Kailola et al., 1993
Sebastidae	<i>Helicolenus dactylopterus</i>	50.0	43*	4.0	Bathydemersal	Allain and Lorange, 2000
Sciaenidae	<i>Argyrosomus regius</i>	230.0	42	4.3	Benthopelagic	González-Quirós et al., 2011
Moridae	<i>Mora moro</i>	80.0	39	3.8	Bathypelagic	Sutton et al., 2010
Pomacanthidae	<i>Pomacanthus maculosus</i>	61.0	36*	2.7	Reef-associated	Grandcourt et al., 2004
Sparidae	<i>Dentex dentex</i>	100.0	36	4.5	Benthopelagic	Baudouin et al., 2016
Sparidae	<i>Diplodus cervinus</i>	55.0	35	3.0	Reef-associated	Jarzhombek, 2007
Lutjanidae	<i>Lutjanus argentimaculatus</i>	150.0	31*	3.6	Reef-associated	Fry et al., 2006
Sciaenidae	<i>Sciaena umbra</i>	70.0	31	3.8	Demersal	Chater et al., 2018
Caproidae	<i>Capros aper</i>	30.0	30	3.1	Demersal	Hüssy et al., 2012
Moronidae	<i>Dicentrarchus labrax</i>	103.0	30	3.5	Demersal	Kottelat and Freyhof, 2007

\*Data for specimens for locations other than Mediterranean Sea.

bass, *Dicentrarchus labrax* can attain age of 30 years (Kottelat and Freyhof, 2007). The species mentioned in this paragraph are economically interesting, and EU Mediterranean countries collect relevant landing and biological data for them [data collection framework (DCF); Regulation (EU), 2017]. It is highly likely, either within monitoring programs or scientific research projects, that otoliths of these species are archived during several years or even decades by different institutions and could be used to extend time series data beyond the maximal reported age.

According to the data presented above, the availability of otoliths for long living species from the Mediterranean is quite limited, as there are only several species reaching a maximum reported age of over 30 years. However, development of statistical techniques enables construction of growth chronologies for shorter living fish species (<15 years) when samples are collected over several years or decades (Coulson et al., 2014). It is possible that, for certain fish species, adequate replicates for chronology construction can be obtained through archive collections.

## OPPORTUNITIES FOR OTOLITH CHEMISTRY RESEARCH

Clarity of growth rings in otoliths is one of the main factors contributing to sclerochronology research, both for growth increment measurements and for otolith chemical analysis (Campana, 1999). Problems related to interpretation of increments in otolith, including age estimation and validation of periodicity, has been pointed out in number of studies in different parts of the world (e.g., Morales-Nin et al., 2005b; Stransky et al., 2005; Hüssy et al., 2016). This problem should not be underestimated, and interpretation of otolith increments needs to be carefully checked and validated. One of the most appreciated characteristics of otoliths is their lack of resorption. This means that once the material has been deposited, the organism will not use these minerals again, even in periods of starvation. Lack of resorption is not shared with other calcified structure (like scales and bones) in fishes or other vertebrates (Bilton, 1974; cited by Campana and Thorrold, 2001). Another special characteristic of

otoliths is that they grow continuously throughout the lifetime of the fish (Campana, 1999).

In order to assign relevant chemistry data to a specific calendar year, it is crucial to distinguish growth increment boundaries (Black et al., 2005; Martino et al., 2019). However, many the most commercially important fish species living in the Mediterranean Sea do not have clearly distinguished growth patterns in their otoliths, which presents a challenge for this type of research. For example, it is still difficult to determine the growth boundaries for the first growth increments in otoliths of *Mullus barbatus* due slow growth and number of false-growth increments laid down before the annulus (Carbonara et al., 2018) and of *Merluccius merluccius* due to the fast growth (de Pontual et al., 2003; Piñeiro et al., 2007; Mellon-Duval et al., 2010) and long spawning period of the species (Morales-Nin and Aldebert, 1997) although a number of direct methods to validate age assessment were used, like mark-recapture (de Pontual et al., 2003; Mellon-Duval et al., 2010), first ring appearance (Belcari et al., 2006), or bomb radiocarbon dating (Vitale et al., 2016).

Species from the families Sparidae and Lutjanidae have annual growth rings, that although thin, are clearly visible (Pidcocke et al., 2015; Winkler et al., 2019), and represent the most promising target taxa for sclerochronology studies. Interesting target species of Sparidae include *Dentex dentex*, *Diplodus cervinus*, and *Sparus aurata*. The latter species, together with *Dicentrarchus labrax* (Moronidae), are particularly interesting, as these are the most important fish aquaculture species throughout the Mediterranean region (Lacoue-Labarthe et al., 2016). In addition to these species and those listed in **Table 1**, another interesting taxon for chemical research of otoliths is *Seriola dumerili* (Carangidae), a species with a circumglobal distribution (Smith, 1997).

Instrumental restrictions, related to the quantity of material required for the analysis of stable isotopes in otoliths, has been the main limitation for the development of isotope related research in otoliths (Sreemany et al., 2017). Due to the small size of the otolith, this resulted in time averaging of data, and analyses were limited to whole otoliths (e.g., Rooker et al., 2008a), or certain parts of otoliths, e.g., the core (Siskey et al., 2016;

Rooker et al., 2019) or edge (Hidalgo et al., 2008; Tanner et al., 2012), without the possibility of obtaining time series data. Development of instruments and methods, including high-resolution laser ablation systems (e.g., Sreemany et al., 2017) and continuous flow isotope ratio mass spectrometry system for ultra-microvolume carbonate samples (Kitagawa et al., 2013; Sakamoto et al., 2017), have opened new opportunities for obtaining time series data from otoliths. Although significant progress was made in instrument development, many are still available only to a limited number of scientists. Therefore, new opportunities for collaborations and research directions related to the Mediterranean are required.

## CHALLENGES AND LIMITATIONS FOR FISH SCLEROCHRONOLOGY RESEARCH IN THE MEDITERRANEAN SEA

Alongside the constraints imposed by the biological characteristics of species, there are other challenges to conducting sclerochronology research in the Mediterranean region. Although otoliths are small structures that are easily archived, there still appears to be a general lack of understanding and effort to construct and manage otolith collections. Panfili et al. (2002) clearly indicated the importance of archiving otoliths, highlighting the need to evaluate, catalog, and conserve otolith collections in a way that will make both the otolith and corresponding fish life history information more accessible to all researchers. The Instituto de Ciencias del Mar-CSIC (Spain) maintains an otolith reference collection that includes samples from different parts of the world, including the Western Mediterranean Sea<sup>2</sup>. One example of an online searchable database of otolith collections from other parts of the world is the otolith collection database housed at the Burke Museum<sup>3</sup>. Although English is generally accepted as a global scientific language, the Mediterranean is highly politically, economically, cultural, and linguistically diverse region, which impacts sample, data, and knowledge storage and sharing. While online searchable otolith collections are not currently possible for a number of reasons, efforts should be made by different institutions or even scientists themselves to archive otoliths together with relevant collection and biology data. It is highly recommended that collections contain samples for different species, not only commercially most important ones, and that special efforts are made to archive otoliths of rare species. Furthermore, Disspain et al. (2016) pointed out the potential to use otoliths from archeological sites to analyze changes in the environment occurring through human history. Linking archeologists with fish biologist and environmental scientists can provide great potential for sclerochronology research.

In addition to the issues related to the availability of otolith collections, continuous sampling over several decades is also a challenge given the limited availability of funding for long-term

studies and the logistics associated with field sampling. Long-term time series data are needed to estimate the real status of exploited resources and their evolution over time (Battaglia et al., 2010) and to analyze climate change effects on marine species and communities (Azzurro et al., 2019). Today, most scientific research projects are short in duration, resulting in difficulties related to securing funds needed for maintaining a longer data time series (Leonart and Maynou, 2003; Rochet and Trenkel, 2003). Even when data collection takes place over longer periods, it often suffers from inconsistencies in sampling design or sampling methods (Rochet and Trenkel, 2003; Rochet et al., 2005). Sampling designs often tend to be incomplete, lacking either randomization or replication, and as such can never conclusively demonstrate the causes of the observed changes. Sampling and storage protocols are often specific to the institution or project. It is, however, encouraging that all the EU Member States bordering the Mediterranean Sea, eight in total, are required to collect fisheries data using unified methodology [Regulation (EU), 2017].

Human resources present another important segment in the development of all marine-related research in the Mediterranean, including sclerochronology. Although otoliths have been analyzed in relation to age and growth, very few attempts have been made to link the data derived from otoliths with environmental data. Education in methods associated with growth increments analysis, chemical composition of otoliths, and statistical methods related to sclerochronology research is strongly needed, either through personal or workshop-based interactions, to stimulate the involvement of fisheries scientists in sclerochronology studies. Furthermore, sclerochronology includes biological, chemical, and physical aspects, requiring an interdisciplinary research team.

Different types of instrumentation are required for sclerochronology analysis. Some instruments need to be readily and continuously available, such as saws and grinding and polishing machines, while those for chemical analysis of otoliths can be offsite. For example, the Laser Ablation System coupled with a High-Resolution Inductively Coupled Plasma Mass Spectrometer (HR-ICPMS) is a sophisticated tool for analysis of elemental composition that is both very expensive and requires specially trained personnel. Careful sample preparation and establishing collaboration with institutions possessing such an instrument can enable the processing of otolith samples at a reasonable cost. Efforts should be made to develop human and research capacities within the framework of different international projects, thereby promoting scientific collaboration and the education of young researchers.

## CONCLUSION

The Mediterranean Sea is a hotspot of marine biodiversity and is also one of the most impacted ecoregions globally (Halpern et al., 2008; Costello et al., 2010), due to increasing levels of human threats that affect all levels of biodiversity (Mouillot et al., 2011; Coll et al., 2012; Micheli et al., 2013) and due to severe impacts from climate change (Lejeune et al., 2010) and biological invasions (Katsanevakis et al., 2012).

<sup>2</sup>ipt.vliz.be/eurobis

<sup>3</sup>www.burkemuseum.org



Determining historical changes in marine communities and consequently fisheries (Pauly and Zeller, 2016) allows us to better understand the present, in order to anticipate the future. This is particularly important in relation to the decline of marine resources (Bell et al., 2017). Thus, it is necessary to develop methods to document long-term trends and detect potential stressors. However, establishing causal relationships between a wide range of stressors and effects at the individual, species, or community level in marine ecosystems is a difficult task that requires the use of multiple lines of evidence (Adams, 2005). Fish are excellent candidates for the study of the effects of climate variability (Pörtner and Peck, 2010). In the Mediterranean Sea, besides the phenomenon of a northward shift in population distribution by native Mediterranean species, the arrival of alien species is also playing an important role in carving the faunal assemblages of the Mediterranean Sea. It is presumed that the coldest parts of the Mediterranean Sea (Gulf of Lyon and North Adriatic) could initially serve as a sanctuary for cold-temperate species, though continued warming of these areas could turn them into a cul-de-sac for such species. This is especially important for endemic species that could become extinct due to the trapping effect (Ben Rais Lasram et al., 2010).

Sclerochronology research has the potential to provide insight into environmental changes in the Mediterranean, both at the local and regional scales (Peharda et al., 2019a). Recent research conducted on bivalves in the Adriatic Sea resulted in a construction of bivalve chronologies (Peharda et al., 2018, 2019a) and geochemical analysis of shells (Markulin et al., 2019; Peharda et al., 2019b), providing important time-series data for comparative analysis and a multispecies approach.

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- Such a multispecies approach has been very promising in other parts of the world, including the work by Black (2009), who analyzed growth increments in trees, bivalves and fish to identify climate variability signals. Further development of fish sclerochronology research in the Mediterranean could facilitate a multi-taxa approach, enabling us to gain a better understanding of environmental drivers in marine habitats.

## AUTHOR CONTRIBUTIONS

SM-S and MP analyzed the data and existing literature in collaboration with DV, HU, and KM. SM-S and MP wrote the draft of the manuscript. All authors conceived the research, and participated in the improvement and revision of the document.

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## SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: <https://www.frontiersin.org/articles/10.3389/fmars.2020.00195/full#supplementary-material>

**TABLE S1** | Published sclerochronology research relating to otolith analysis in the Mediterranean Sea.

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**Conflict of Interest:** The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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