



Comparing early life traits of hakes from Chilean Patagonian fjords inferred by otolith microstructure analysis



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ABSTRACT

Comparison of larval growth and back-calculated hatch dates on the basis of microstructure analysis of sagitta otoliths were carried out in wild-caught larvae of three commercially important gadiform fishes from Northern Patagonia, South Pacific, Chilean hake *Merluccius gayi*, southern hake *Merluccius australis*, and Patagonian grenadier *Macruronus magellanicus*. Because of the morphological similarities in their larval stages, we hypothesize that early life traits (ELT) will be different among the hake species in order to avoid/reduce interspecific competition. During October–November 2011, there was differences in the spatial distribution of the surviving larvae from the three species along the inner sea; while larval Chilean hake were concentrated in the warmer and fresher seawaters of the Reloncaví fjord, larval Patagonian grenadier were aggregated southward, in the Ancud Gulf, while larval southern hake were found throughout the area, but in low density. Temporal differences were evident in the back-calculated hatch days among species; larval hakes of genus *Merluccius* hatched during late October, while larval grenadier hatched between August and September. Estimated larval growth rates differed among hake species; larval *M. magellanicus* experienced the fastest growth rates ($0.334 \pm 0.01 \text{ mm d}^{-1}$), while larval *M. australis* experienced growth rates of $0.220 \pm 0.01 \text{ mm d}^{-1}$, and larval *M. gayi* showed the slowest growth rates ($0.180 \pm 0.01 \text{ mm d}^{-1}$). Therefore, resource partitioning both in time and space are occurring among early life stages of *M. gayi*, *M. australis* and *M. magellanicus* during austral spring season in Northern Patagonian fjords of Chile.

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1. Introduction

Early life traits (ELT), like larval growth rates and spawning/hatching dates, have been proven to be crucial features in improving our understanding of processes potentially affecting recruitment and subsequent life stages (Contreras et al., 2013; Di Franco et al., 2013). Otolith microstructure analysis represents a powerful tool for estimating individual age, growth rate, and hatch dates in larvae and juveniles (Campana, 2005; Grote et al., 2012a; Palacios-Fuentes et al., 2014). In particular, the correlation between

otolith size and somatic size enables growth trajectories and histories to be back-calculated (Takasuka et al., 2008).

Also, otoliths of fish larvae may provide information on biophysical interactions such as the effects of low-salinity plumes in coastal waters (Landaeta et al., 2012), ocean acidification (Munday et al., 2011), vertical mixing and ice melting (Zenteno et al., 2014); the information obtained through the analysis of daily fish otoliths microstructure has been a critical component of fisheries management and of ecological and oceanographic processes (Sponaugle, 2010).

The outer and inner seas of Western Patagonia, South Pacific, are utilized as spawning and nursery areas of several marine ground-fish resources, such as the southern hake, *Merluccius australis* and Patagonian grenadier, *Macruronus magellanicus* Lönnberg, 1907 (Balbontín et al., 2004; Bustos et al., 2008a,b; Schuchert et al., 2010). Southern hake is a demersal fish species of the southern

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hemisphere distributed along the coast of Argentina 50–55°S associated with the cold waters of the Malvinas Current, and in southern Chile from 38 to 55°S (Cousseau and Perrotta, 1998). This species main spawning area is located along the shelf break and canyons in the northern area of Chilean Patagonia, close to Guafo and Guambelin Islands (43–45°S), and spawning occurs in austral winter from July to September (Balbontín and Bravo, 1993). Nonetheless, in the last decade, a large number of eggs and larvae have also been reported in spring in some inshore areas of the northern Patagonia (Bustos et al., 2007; Castro et al., 2011).

Patagonian grenadier is a eurybathic species that exhibits strong affinity for Subantarctic waters (SSAW) (Cousseau, 1993). It inhabits depths from 30 to 500 m depth on the outer part of the continental shelf (Machado-Schiaffino and Garcia-Vasquez, 2011). The largest known spawning aggregations occur between 44 and 47°S, in submarine canyons located on the Pacific shelf, and Niklitschek et al. (2014) suggest that the proximity of principal spawning area to the bifurcation of the West Wind Drift Current may cause large inter-annual and decadal variations in larval transport and the relative contribution of different shelf and Patagonian Inner Sea nurseries to recruitment.

In the last decade, large abundance of South Pacific hake *Merluccius gayi* eggs and larvae have been found in the inner sea of Northern Patagonia, mainly in the first 50 m depth of the water column of the Reloncaví fjord (Bustos et al., 2008b; Landaeta et al., 2009). This suggest that the area is currently used as spawning and early nursery ground, with egg and larval densities as large as those occurring in spawning grounds off central Chile (Vargas and Castro, 2001; Landaeta and Castro, 2006, 2012). Until date, all ecological and fishery information regarding South Pacific hake spawning grounds, larvae (Vargas and Castro, 2001; Landaeta and Castro, 2006), juveniles (San Martín et al., 2011) and adults (San Martín et al., 2013) is on the basis of studies carried out in upwelling-influenced areas, but none is currently known about early life traits of the stock inhabiting fjord ecosystem of Pacific Patagonia.

The inner sea area is characterized by a high degree of geomorphologic and hydrographic complexity, forming one of the largest estuarine areas in the world, as well as strong climatic seasonal fluctuations (i.e., solar radiation and precipitation), partially modulating the seasonal changes in phytoplankton biomass and primary production (González et al., 2010). Chlorophyll *a* (Chl-*a*) ranges in this inner sea correspond to those values found for highly productive zones, such as coastal upwelling areas (0.05–30 mg Chl-*a* m⁻³, Lara et al., 2010). In this area, large abundance of copepod eggs, nauplii and copepodite as well as the large water column stability in surface waters may promote larval feeding success (as it has been detected in other fish larvae of the region, Landaeta et al., 2011, 2013) and faster larval growth rates in Gadiform species that inhabit surface waters of Patagonian fjords (Castro et al., 2011).

In this study, growth and hatch dates of field-caught larvae of South Pacific hake, *M. gayi*, southern hake, *M. australis* and Patagonian grenadier *M. magellanicus* were analyzed to investigate interspecific variations of ELT in an area where all these species coexist. There is no previous information about larval growth for neither of three species for Chilean waters. Because of the morphological similarities in their larval stages, such as preanal length, snout length, and eye diameter (Balbontín et al., 2004), we hypothesize that ELT will be different among the hake species to avoid/reduce interspecific competition. Therefore, the objective of this study is to estimate and compare larval growth and back-calculated hatch dates, on the basis of otolith microstructure analysis, from three commercially important gadiform fishes from Northern Patagonia, South Pacific.

2. Methods

2.1. Study area

The inland sea of southern Chile features several areas of steep bathymetric and morphologic change where tidal (Valle-Levinson et al., 2001; Cáceres et al., 2006) or wind forcing (Valle-Levinson and Blanco, 2004) can substantially modify the two-layer exchange observed elsewhere. The temporal variability of the flow in the region is dominated by the semidiurnal tide (Molinet et al., 2006). For example, currents in the Ancud Gulf measured during spring 2004, shows presence of internal waves of significant magnitudes (about 40 cm s⁻¹) that may promote entrainment to upper layers and zooplankton transport into the deepest layers; also, the sea level and currents at 100 m depth seems to be out of phase by about 3 h (Castro et al., 2011).

2.2. Field work

Fish larvae were collected during an oceanographic cruise carried out during 17th October to 15th November 2011 in the inner sea of Chiloé, Northern Patagonia, Chile (Fig. 1). A north-south transect was sampled, comprising 20 stations. At each sampling station, a Seabird SBE-19 CTD cast from surface to 200 m depth was carried out. Then, plankton samples were obtained in 17 of the 20 stations (Fig. 1) with oblique hauls from surface to 100 m depth by using a 1 m² Tucker trawl equipped with 300 μm nets and a General Oceanics flow meter mounted in the frame of the net, to estimate the filtered seawater. Two samples were taken during each trawl, 0–100 m and 100–0 m depth. All plankton samples were initially preserved with 4% formalin buffered with sodium borate, and then they were preserved in 96% ethanol.

2.3. Laboratory work

A total of 106 ichthyoplankton samples were analysed. Larvae of *M. gayi*, *M. australis* and *M. magellanicus* were identified following Balbontín et al. (2004), and Bustos and Landaeta (2005). Larval densities were expressed as individuals (ind.) 1000 m⁻³. The developmental stages of all larvae were classified as preflexion (yolk-sac larvae were included in this group) or postflexion (flexion and postflexion larvae were pooled together). In addition, the body lengths (BL) of the undamaged larvae (*M. gayi* = 317, *M. australis* = 20, *M. magellanicus* = 79) were measured (notochord length, NL or standard length, SL) to the nearest 0.01 mm under an Olympus SZ-61 stereomicroscope (Olympus Corporation, Shinjuku-ku, Tokyo, Japan) using a Moticam 2500 (5.0 Mpixel) video camera (Motic Instrument, Inc.; Richmond, BC, Canada) that was connected to a PC containing the Moticam Image Plus 2.0 software (Motic China Grup, Co.; Xiamen, China). The larval measurements were not corrected for shrinkage.

The left and right sagittae otoliths were removed using dissecting needles from 90 randomly selected larval *M. gayi*, 18 larval *M. australis* and 64 larval *M. magellanicus*. The otoliths were embedded in epoxy resin on a glass slide. Age was estimated by counting the number of daily increments from a dark prominent increment (the hatch check, HC, Alvarez and Cotano, 2005) to the otolith edge using a Motic BA310 light microscope (Motic Instrument, Inc.; Richmond, BC, Canada) at 1000× magnification under oil immersion. The hatch mark has been validated for otoliths of *Macruronus novaezelandiae* (Thresher et al., 1988). The increments were assumed to be formed daily according to Bailey (1982), Thresher et al. (1988) and Morales-Nin et al. (2005). An image analysis software (Moticam Image Plus 2.0) was used to obtain the mean values, resulting in three independent measurements, for both the longest radius, perimeter and area of the sagitta.

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