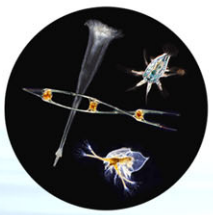
Journal of
Plankton Research

academic.oup.com/plankt



J. Plankton Res. (2018) 40(5): 606–618. First published online August 28, 2018 doi:10.1093/plankt/fby033

Larval distribution and connectivity of the endemic Sciaenidae species in the Upper Gulf of California

Y. GARCÉS-RODRÍGUEZ[1], L. SÁNCHEZ-VELASCO[1]*, N. DÍAZ-VILORIA[1], S. P. A. JIMÉNEZ-ROSENBERG[1], V. GODÍNEZ[2], J. MONTES-ARECHIGA[3] AND A. MUNGUIA-VEGA[4]

[1]INSTITUTO POLITÉCNICO NACIONAL–CENTRO INTERDISCIPLINARIO DE CIENCIAS MARINAS (IPN-CICIMAR), AV. IPN S/N, LA PAZ, B.C.S. 23096, MEXICO, [2]CENTRO DE INVESTIGACIÓN CIENTÍFICA Y DE EDUCACIÓN SUPERIOR DE ENSENADA (CICESE), 3918 ENSENADA-TIJUANA ROAD, ZONA PLAYITAS, ENSENADA, B.C. 22860, MEXICO, [3]DEPARTAMENTO DE FÍSICA, UNIVERSIDAD DE GUADALAJARA, AV. REVOLUCIÓN 1500, COL OLÍMPICA, C.P. 44440 GUADALAJARA, JALISCO, MEXICO AND [4]CONSERVATION GENETICS LABORATORY, SCHOOL OF NATURAL RESOURCES AND THE ENVIRONMENT, THE UNIVERSITY OF ARIZONA, TUCSON, AZ, USA

*CORRESPONDING AUTHOR: lsvelasc@gmail.com

Received March 15, 2018; editorial decision July 30, 2018; accepted August 2, 2018

Corresponding editor: Xabier Irigoien

Because the endemic Sciaenidae species (commonly known as drum or croakers) are important to the fishing industry in the Upper Gulf of California, their larval distribution and connectivity was analyzed in relation to hydrographic conditions during spawning periods (March, June and September). *Totoaba macdonaldi*, *Micropogonias megalops*, *Menticirrhus nasus* and *Cynoscion othonopterus* larvae were morphologically and genetically identified. Genetic analysis reveals for the first time the presence of *Isopisthus remifer*, which had not previously been morphologically identified. The most relevant hydrographic structure in the region was the permanent stratification front ($\Phi = 10\,J/m^3$) originated by the convergence of mixed water of the Northern Gulf (~20 m depth) and stratified water coming from the adjacent oceanic water (~200 m). Whereas *T. macdonaldi* larvae were only collected in the shallowest area in March, the other species were found mostly along the front in June and September. Connectivity matrixes showed high particle retention along the front (>80%) coinciding with the larval distribution. Results indicate that the stratification front might favor larval survival and prevent their advection toward the ocean. This type of retention likely enhances endemism, not only of these species but also of others coastal demersal species.

KEYWORDS: Fish larvae; Sciaenidae species; *Totoaba macdonaldi*; distribution; larval connectivity; Upper Gulf of California; Biosphere Reserve

available online at academic.oup.com/plankt

INTRODUCTION

Management and conservation of marine reserves is a complex task, since it involves defining and coupling physical and biological interactions at multiple spatial and temporal scales, as well as anthropogenic factors (Cudney-Bueno *et al.*, 2009; Pollnac *et al.*, 2010). Management actions are often established using a cautionary approach, without in-depth knowledge of relevant factors, such as the spawning strategies (areas, periods and intensity) and relationships with physical environmental conditions of focal species (Borguez *et al.*, 2009; Cudney-Bueno *et al.*, 2009), that may affect management efficacy.

For example, the Upper Gulf of California (UGC), located in the north of the Gulf of California (Fig. 1), was declared a Biosphere Reserve in 1993 (DOF, 1993). This was due in large extent to the presence of endemic fish species, mostly of the Sciaenidae family, such as *Totoaba macdonaldi*, *Micropogonias megalops*, *Cynoscion othonopterus* and the endemic and critically endangered porpoise, the Vaquita *Phocoena sinus* (Rojas-Bracho *et al.*, 2006). In addition, it is an area of breeding and development of many species of commercial and ecological importance, such as shrimp (*Litopenaeus stylirostris*) (Hastings *et al.*, 2010; Rodríguez-Quiroz *et al.*, 2010). However, there are national and international controversies about fishery management of some fishes and shrimps and the Vaquita conservation, and there is a critical need for information on the life cycle of species of ecological and commercial importance to have informed management (Cisneros-Mata *et al.*, 1995; All, 2006; Rodríguez-Pérez *et al.*, 2018).

The UGC has a shallow shelf (<30 m depth) with irregular bathymetry where depth increases to the Wagner Basin (Lavín *et al.*, 1998). It is a highly seasonal and macro-tidal inverse estuary, with temperature ranging from ~14°C in winter to 32°C in summer (Lavín *et al.*, 1998). The seasonal circulation is cyclonic most of the year, with maximum speed (~0.20 m/s) in June, while in the winter months, there is no well-defined pattern of circulation and the currents are weak (~0.03 m/s) (Montes *et al.*, 2015). The UGC is vertically well mixed throughout the year (Lavín *et al.*, 1998). The strong mixture caused by the tidal current generates a stratification front that delimits the well-mixed shallow areas of the UGC from the deep and stratified areas of the northern Gulf of California (Argote *et al.*, 1995). This stratification front (with temperature, salinity, chlorophyll *a* and dissolved oxygen concentration gradients) is a permanent feature and is located on the southern edge of the UGC, with seasonal variations in its position. During the summer, this front is close to the 30 m depth contour and in winter it is positioned over the 60 m depth contour (Argote *et al.*, 1995; Lavín *et al.*, 1998). The influence of this front on the species that inhabit the UGC has not been described to date.

It is assumed that *T. macdonaldi* spawns from February to April with a peak in March (Cisneros-Mata *et al.*, 1995), and that *C. othonopterus*, *Menticirrhus nasus* and *M. megalops* spawning occurs from February to September with maximum intensity in summer (Erisman *et al.*, 2012; Sadovy and Erisman, 2012). Given that the *T. macdonaldi* species is classified as an endangered species (NOM-059-SEMARNAT-2010), commercial fishing has been restricted. However there is a great black market for swim bladder of this species in China (Anonymous, 2016b), and it has been difficult for governmental institutions to curtail the illegal exploitation. Further *M. megalops* and *C. othonopterus* support important fisheries in the UGC, although there is little clarity in their fishery managements due to the lack of solid information on their biology (Valenzuela-Quiñonez *et al.*, 2014); and even less is known of their planktonic stages (Sánchez-Velasco *et al.*, 2012).

Previous studies in the UGC reported six larval morphotypes in the Sciaenidae family located mainly in the shallowest and saltiest zone in the north of the UGC (Sánchez-Velasco *et al.*, 2012). The molecular identifications of Sciaenidae larvae in this region started with the Díaz-Viloria *et al.* (2013) and Camacho-Gastélum *et al.* (2016) studies, through mitochondrial DNA (mtDNA) markers. These authors identified the young larvae of *T. macdonaldi*, *C. othonopterus*, *Cynoscion reticulatus*, *Me. nasus* and *M. megalops*. These kinds of studies opened the possibility to understand the aspects of the life cycle of these Sciaenidae species in the UGC. Although, to date, there is no certainty about basic but fundamental issues such as where these species spawn, which oceanographic conditions could favor larval survival and the factors that have contributed to their endemism.

We expected to find high larval abundance around the stratification front, possibly associated with retention processes. The stratification front might also prevent larval dispersion outside the UGC. The objective of this study was to describe larval distribution and connectivity of endemic Sciaenidae species (*T. macdonaldi*, *C. othonopterus*, *Me. nasus* and *M. megalops*) in relation to the stratification front in the UGC during early spring (March 2011) and early and late summer (June 2013 and September 2012), which are the main spawning periods of these species. This information will increase our understanding of possible mechanisms contributing to endemism of both these and others coastal demersal species that inhabit this region.

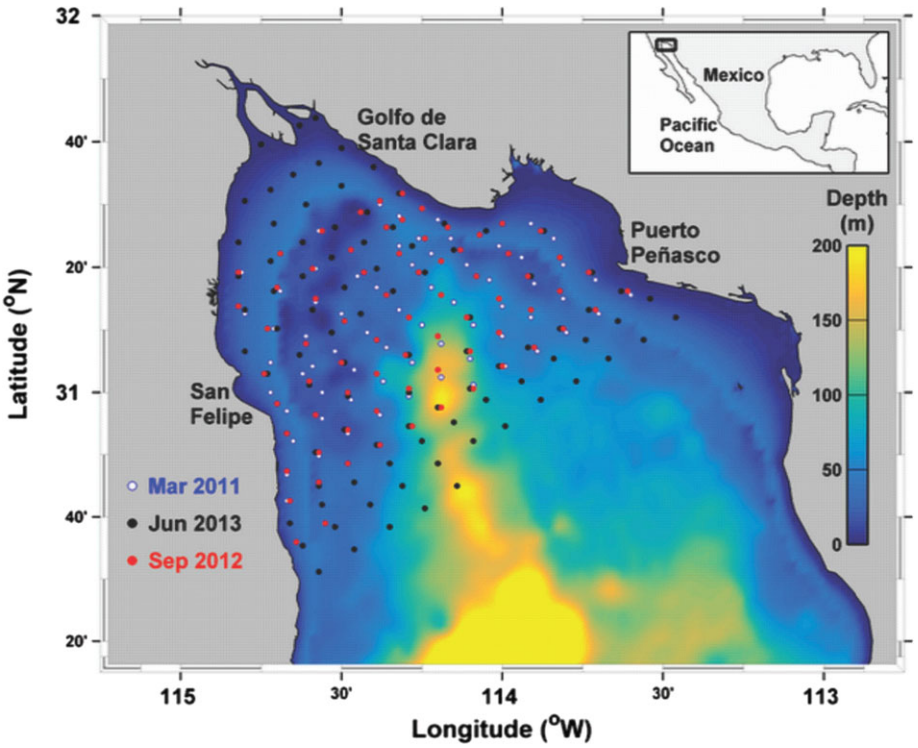


Fig. 1. Map of the UGC with bathymetry (in meters) and the locations of sampling stations.

MATERIALS AND METHODS

Sampling methods and data analysis

Physical data and zooplankton samples were obtained from three oceanographic cruises carried out aboard the R/V Francisco de Ulloa in the UGC, during early spring (March 2011), and early and late summer (June 2013 and September 2012) (Fig. 1). Vertical profiles were obtained at each station using a SeaBird 911plus CTD probe equipped with dissolved oxygen and fluorescence sensors. Conservative temperature (Θ; °C) and absolute salinity (S_A; g/kg) were calculated from *in situ* temperature and practical salinity was calculated with TEOS-10 software (www.TEOS-10.org; IOC *et al.*, 2010).

A measure of the stratification intensity in the water column, the Φ stratification parameter was calculated with the equation proposed by Simpson (1981), which considers the profile of potential density, the average density of the water column, the maximum depth of the integration and the gravitational acceleration. The Φ parameter represents the amount of energy (J/m^3) needed to mix a water column a completely to a given depth. For total mixing of the water column Φ = cero, and for a frontal zone, Φ is ~10 J/m^3 according to Argote *et al.* (1995).

Oblique zooplankton trawls were carried out at each station using opening–closing conical nets (60 cm mouth diameter, 250 cm net length, 0.505 mm mesh), at three different depths (from 15 to 10 m, from 10 to 5 m and from 5 m to the surface). The volume of filtered water was calculated using calibrated flow meters placed at the mouth of each net. The nets were towed in a circular path at 2.5 knots for 5 minutes, during the day or night (Smith and Richardson, 1979). Two samples were obtained at each station, the first was preserved in 5% formalin and the second preserved in 80% ethanol. The first sample was used for larval morphological identification and to determine the larval abundance of species, while the second was employed for genetic corroboration of Sciaenidae species.

Morphological fish larvae identification

Prior to identification, larvae of each species were divided into three developmental stages: preflexion, flexion and postflexion based on the degree of flexion of the terminal section of the notochord during formation of the caudal fin (Kendall *et al.*, 1984). Larvae were classified to the family levels by their morphological characteristics (mainly meristic and pigmentation). Sciaenidae

larvae have 25–26 myomeres (5 pre-anal and 20 postanal myomeres), somewhat bulbous heads, coiled guts and relatively slender, tapering tails (Moser, 1996). Larvae of *C. othonopterus*, *M. megalops*, *Me. nasus* and *T. macdonaldi* were identified to the species level through diagnostics of their pigmentations (Díaz-Viloria *et al.*, 2013; Camacho-Gastélum *et al.*, 2016).

After larval identification, counts were standardized to the number of larvae per $10\,m^2$ of sea surface, according to the procedures established by Smith and Richardson (1979).

Molecular identification of larvae

Nine adult specimens of Sciaenidae: *T. macdonaldi* ($n = 1$), *Cy. reticulatus* ($n = 2$), *Isopisthus remifer* ($n = 3$) and *M. megalops* ($n = 3$) were collected from the eastern part of the UGC (31° 07′ N, 114° 29′W), on 26 January 2016 (Fig. 1). Adults were identified to the species level (Chao, 1995).

Genomic DNA extractions from adults and larvae were conducted using the salt precipitation technique proposed by Aljanabi and Martinez (1997). Two mtDNA fractions were amplified: the cytochrome *c* oxidase, subunit I (COI) of ~700 bp. The COI fraction was amplified with the primers: VF1 (5′-TTCTCAACCAACCACAAAGACATTGG-3′) (Ivanova *et al.*, 2007) and FishR1 (5′-TAGACTTCTGGGTGGCCAATCA-3′) (Ward *et al.*, 2005). DNA amplifications were done in a PCR thermal cycler (Techne, Staffordshire, UK), following the thermal conditions described in Camacho-Gastélum *et al.* (2016). PCR products (20 mL, 100 ng/μL) were sequenced in both directions in an automatic sequencer (ABI Prism 3730XL, Applied Biosystems, Carlsbad, CA, USA) by Macrogen (Seoul, Korea). Twelve COI sequences from six species of Sciaenidae from GenBank were used as references (Table I). Sequences were trimmed to 486 bp to ensure the sequence equality of all individuals. Genetic distances were obtained according to the Kimura 2-parameter model (Kimura, 1980). Adult and larvae sequences were used to display phylogenetic relationships by neighbor-joining (NJ) tree (5000 bootstraps) with MEGA version 6.0 software (Tamura *et al.*, 2013).

Table I: Reference sequences with the GenBank accession number

	COI	
Species	*n*	Access number
Totoaba macdonaldi	1	KC208684
Cynoscion othonopterus	3	KR632704, KR632705, KC208685
Menticirrhus nasus	3	KR632722, KR632725, KR632727
Micropogonias megalops	3	KR632710, KR632719, KC208691
Cynoscion reticulatus	1	KC208680
Cynoscion parvipinnis	1	GU440301

Simulations of larval dispersal

The Delft3D hydrodynamic numerical model (flow module) was used to estimate the speed and direction of the current in the study area. The model that solves the Navier–Stokes equations on a curvilinear mesh, has a grid size of 1 km in the horizontal and 12 levels in the vertical. The physical forces at the open borders included the main components of tide, average climatological data of the North American Regional Reanalysis (Mesinger *et al.*, 2006) and physical data (sea level, temperature, salinity) of Simple Ocean Data Assimilation (Carton and Giese, 2008). We estimated particle trajectories following the advection/diffusion scheme described in Proehl *et al.* (2005) and Visser (2008).

Five zones were delineated in the UGC according to the larval abundance distributions and hydrographic characteristics (bathymetry, circulation patterns and position of the stratification front) (Fig. 2). These sites include the Colorado River Delta zone (AG1), the strong vertical mixing zone (AG2 and AG3) and the frontal zone (AG4 and AG5). Passive particles 6500 were launched in each zone, were integrated for the five sites, and the results were expressed as total export values. We conducted simulations for three launch dates (March 2011, June 2013 and September 2012), which coincide with the spawning season of most Sciaenidae species in the UGC. In each particle release, the simulation time was 7 days, to represent the preflexion larval stage of the Sciaenidae family.

Connectivity matrices were generated using the proportion of particles that were established in each location in relation to the total number of particles released at each site. The probability of local retention (diagonal in the connectivity matrix) was calculated as the proportion of locally produced particles that remained within the spatial unit at the end of the simulation.

RESULTS

Distribution of environmental conditions and stratification front

During March 2011, the conservative temperature (Fig. 3a) showed a range from 16 to 20°C with minimum values (<17°C) in the central part of the UGC and maximum values in the southern part (>18°C). The absolute salinity (Fig. 3b) fluctuated between 35.6 and 36.4 g/kg, decreasing from north to south. The

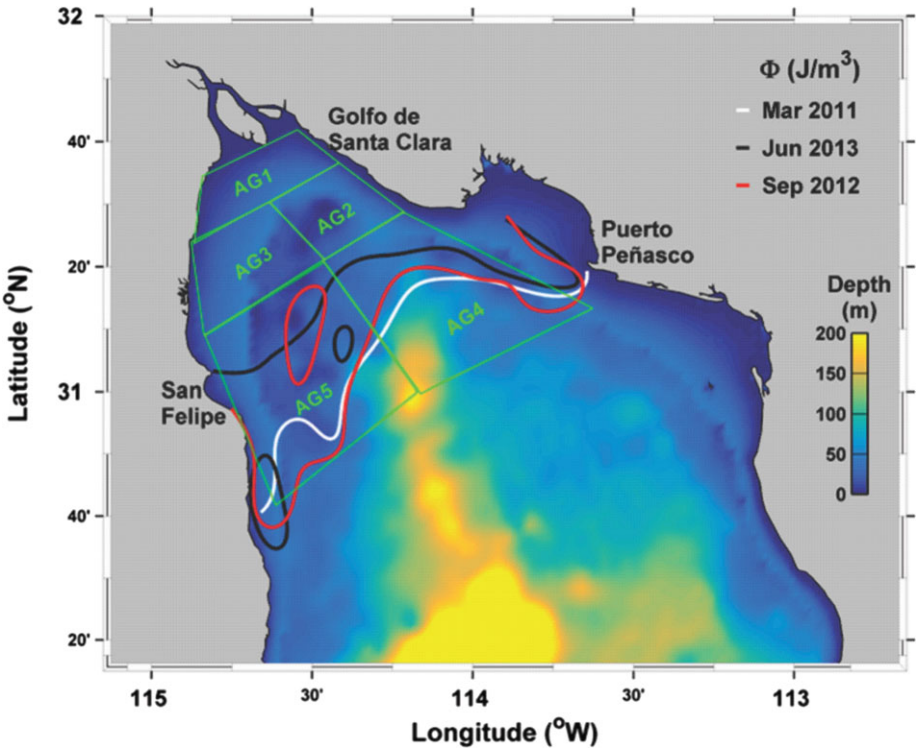


Fig. 2. The solid line (white, black and red) shows the spatial distribution of stratification fronts (parameter $\Phi = 10\,J/m^3$) in the UGC. The squares show the five areas where the particles were sowed to determine the connectivity matrixes.

northwest part of the UGC, i.e. north to San Felipe, was the saltiest zone (>36 g/kg). Dissolved oxygen (Fig. 3c), ranged from 220 to 300 μmol/kg, showed a gradient from south of the study area (300 μmol/kg) to north of San Felipe (220 μmol/kg). Chl *a* concentration (Fig. 3d) did not show a clear pattern, but the highest values were observed to the south of San Felipe (>3 mg/m^3).

During June 2013 sampling, surface temperatures (Fig. 4a) had a wider range (25–30°C) than in March, with minimum values (25°C) in the central and southern part of the UGC and the maximum values (30°C) in the northwest shallow zone. Absolute salinity (Fig. 4b) fluctuated between 35.6 and 36.8 g/kg, with values increasing from south to north, where highest values were in the shallow areas of the UGC (>36 g/kg). Dissolved oxygen (Fig. 4c) ranged from 180 to 250 μmol/kg and an inverse gradient in comparison to the previous variable, with lowest concentrations (180 μmol/kg) in shallow areas to the northern and highest (250 μmol/kg) to the south of Punta Borrascoso. As with salinity, Chl *a* concentrations (Fig. 4d), showed a gradient from south to north with highest values (~1 mg/m^3) observed in the shallow area to the northern of the UGC.

During September 2011, the highest temperature values were observed (Fig. 5a), fluctuating between 31 and 32°C. The minimum values were in the central and southern part of the UGC and the maximum in northwest part of the study area. Absolute salinity (Fig. 5b) fluctuated between 35.8 and 37 g/kg, with lowest values in the south of the UGC (35.8 g/kg), increasing toward the north of San Felipe (37 g/kg). Dissolved oxygen (Fig. 5c), with a range from 180 to 220 μmol/kg, tended to be homogeneously distributed. Chl *a* concentration (Fig. 5d) had the highest values to the north and southeast part of the UGC (~2 mg/m^3).

In March, the stratification front ($\Phi = 10$) was located between San Felipe and Puerto Peñasco (Fig. 2, white line), in the southern part of the UGC at average depth of 50 m, indicating that there were vertical mixing conditions throughout the UGC. In June, the front (Fig. 2, black line) was located more to the north than in March with an average depth of 30 m. In September (Fig. 2, red line), the front had a position similar to March, located between San Felipe and Puerto Peñasco.

Morphological fish larvae identification and genetic corroboration

Nineteen sequences of COI (486 bp) were obtained from adults and larvae and submitted to GenBank. The mean intraspecific genetic divergences within the four

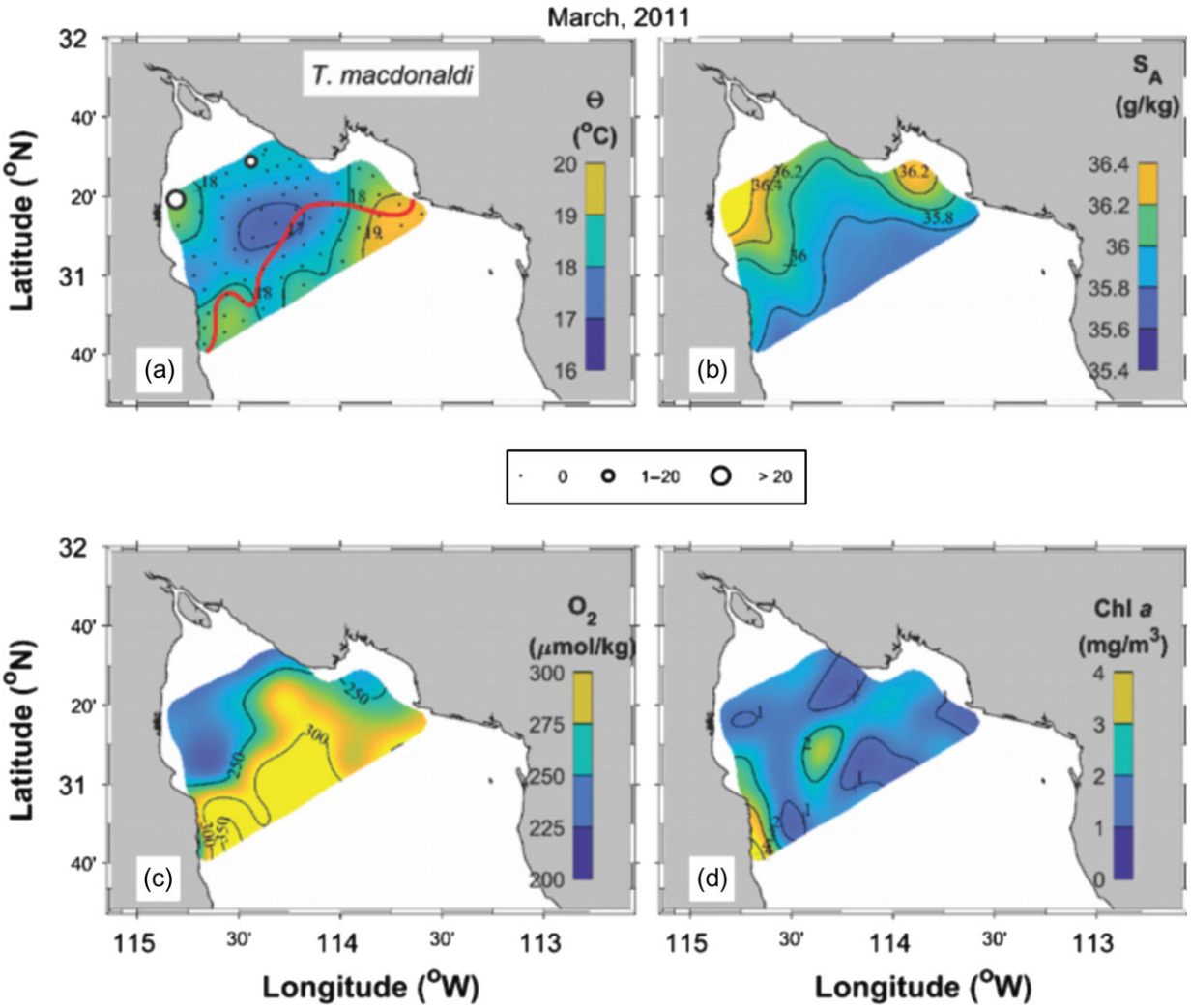


Fig. 3. Surface hydrography for March: (**a**) temperature (°C), (**b**) absolute salinity (g/kg), (**c**) dissolved oxygen (μmol/kg) and (**d**) chlorophyll *a* (mg/m^3). The red contour line indicates the position of the stratification front. The size of the white circle represents the larvae abundance.

species (2–4 adult and 10–12 larvae), were <1% (0.1–0.9%). The mean interspecific genetic divergences among the four species were higher than 2% (15.6–23.3%). The NJ tree showed that six larvae identified as *Me. nasus*, six of *M. megalops*, and three of *C. othonopterus*, were grouped to reference sequences of those species, such groups showed bootstrap support of 100%. Based on genetic divergences and NJ tree COI, the larvae were corroborated to species level.

However, three larvae (two in preflexion and one in flexion), misidentified as *C. othonopterus*, were grouped with sequences of *Isophistus remifer* (Fig. 6). Taking into account the possible misidentification of *I. remifer* larvae like *C. othonopterus*, hereafter the larvae of both species were grouped as *C. othonopterus-I. remifer complex*.

Abundance and distribution of fish larvae

A total of 3115 larvae were identified from the formalin samples (Table II), where most of the larvae (~85%) were collected in the surface strata (0–5 m depths) in preflexion stage (>80%). The highest values of larval abundance was found in June (58%) and September (39%), while the lowest in March (1.6%) (Table II).

Totoaba macdonaldi larvae were only recorded in March in two sampling stations located in the north of the UGC (Fig. 3a), with relativity low larval abundance (Table II). Its presence was associated with the salty and cold water.

In June, larvae of *C. othonopterus-I. remifer complex*, *M. megalops* and *Me. nasus* were identified (Table II). The highest larval abundances were located throughout the

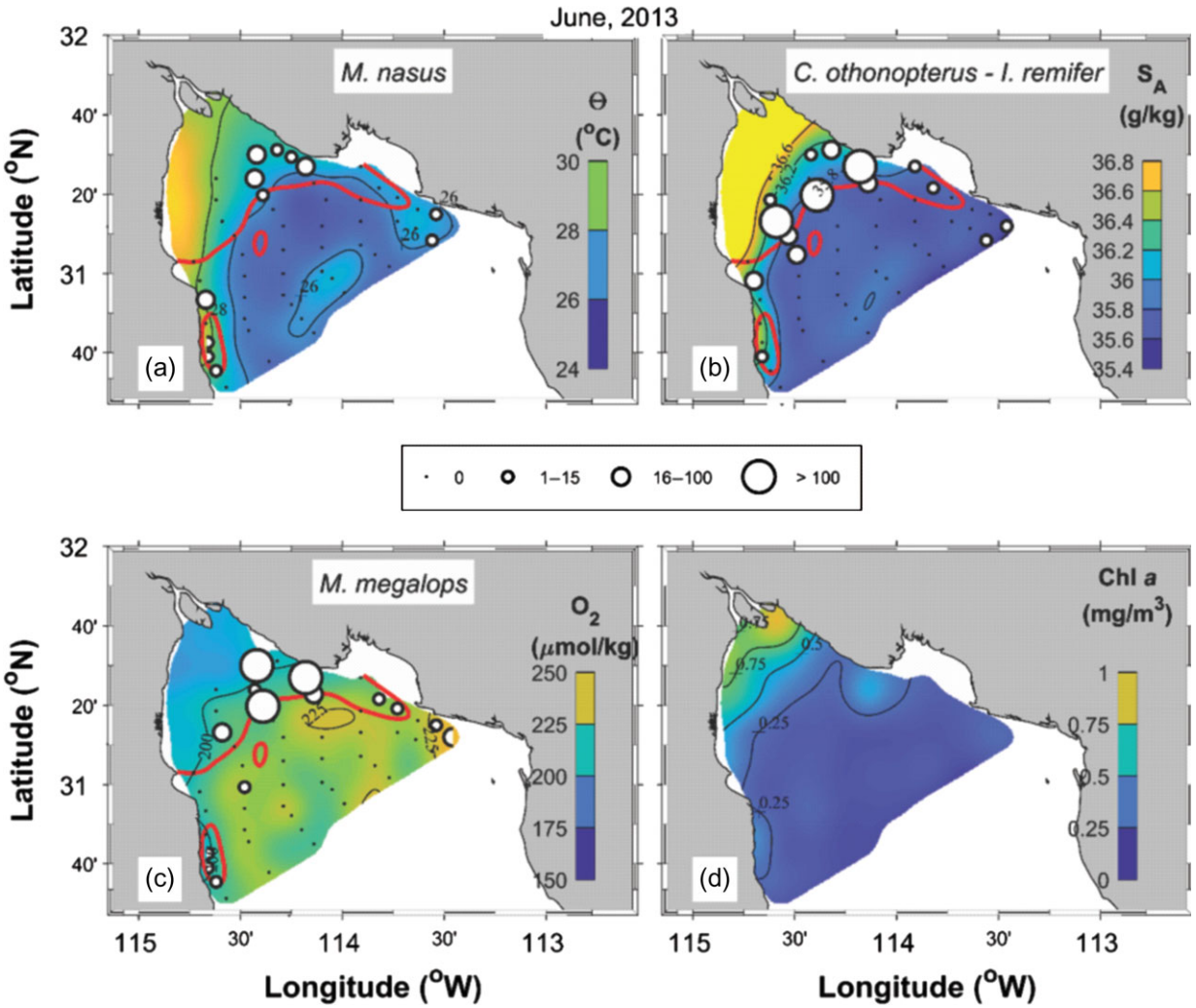


Fig. 4. Surface hydrography for June: (**a**) temperature (°C), (**b**) absolute salinity (g/kg), (**c**) dissolved oxygen (μmol/kg) and (**d**) chlorophyll *a* (mg/m^3). The red contour line indicates the position of the stratification front. The size of the white circle represents the larvae abundance.

stratification front, with major concentrations on its eastern side (Fig. 4a–c).

In September, the larval abundance of *C. othonopterus-I. remifer complex* and *Me. nasus* increased, while the larval abundance of *M. megalops* decreased (Table II). The larvae of *C. othonopterus-I. remifer* complex and *Me. nasus* showed a wide distribution around and to the north of the stratification front. The *M. megalops* larvae had the same distribution, but with the lowest abundance (Fig. 5a–c).

Fish larval connectivity

The connectivity matrices (Fig. 7) of the five zones defined here for the UGC (Fig. 2) showed the highest particle retention along the quadrants that covered the stratification front (AG4 and AG5). In March (Fig. 7a), the greatest particles' retention associated with the frontal zone was observed, with values near to 90% of retention. While the 10% of the remaining particles were transported to the mixed zone (AG2 and AG3). The delta zone (AG1) showed relativity high retention (~60%) and transport of remaining particles towards the mixed zone (40%). The lowest particle retention was observed in the mixed zone, with values near 40%, while the rest of the particles (60%) were transported towards the front zone.

Particles retention decreased slightly in June and September (Fig. 7b, c) with respect to March, but the connectivity pattern was similar to March. The front zone showed the highest retention of particles (70% in June and 80% in September) with transport towards the mixed zone (30 and 20%). The delta zone displayed a particle retention near 60% in both periods and the rest

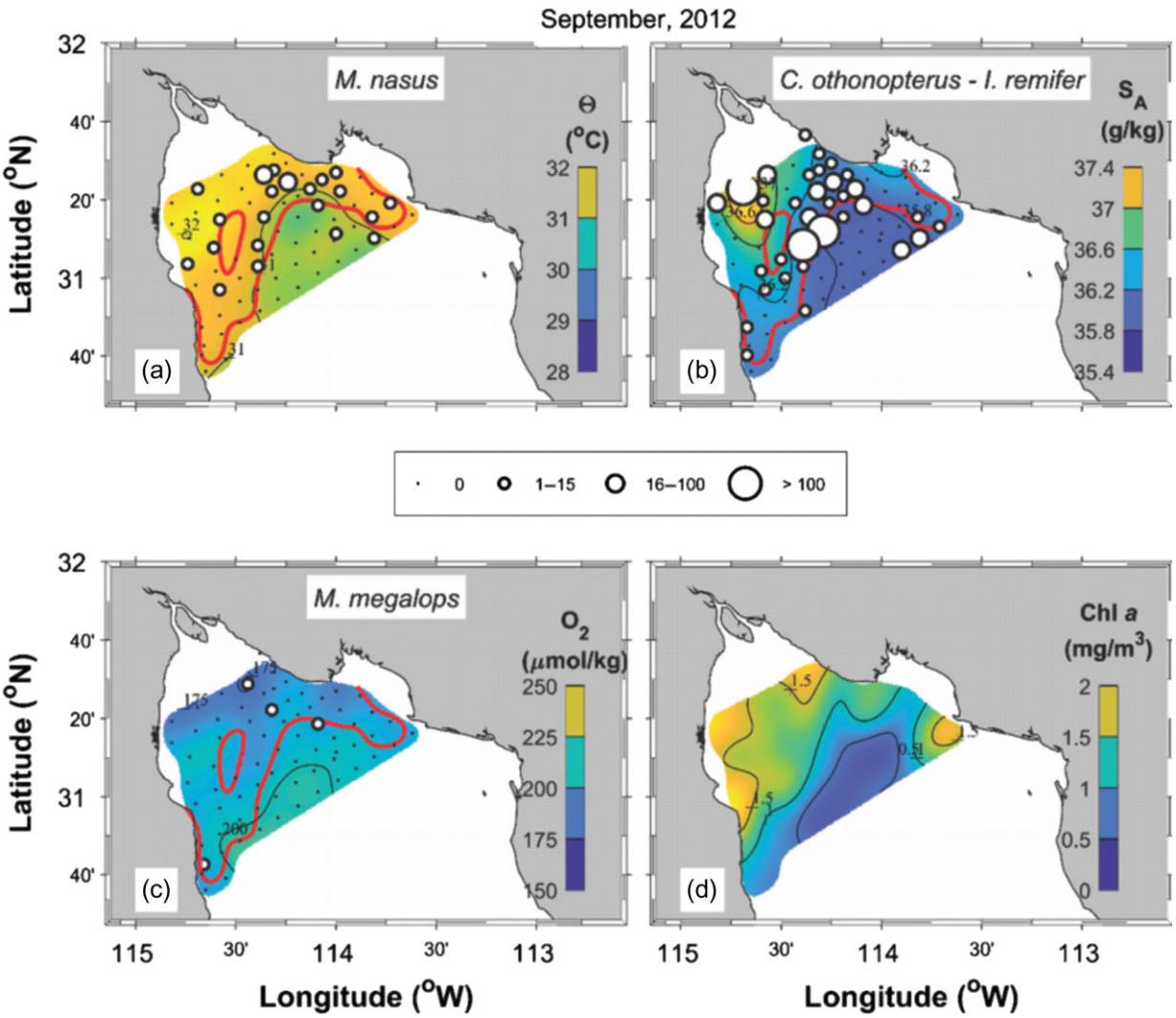


Fig. 5. Surface hydrography for September: (**a**) temperature (°C), (**b**) absolute salinity (g/kg), (**c**) dissolved oxygen (μmol/kg) and (**d**) chlorophyll *a* (mg/m^3). The red contour line indicates the position of the stratification front. The size of the white circle represents the larvae abundance.

of the particles (40%) were transported to the mixed zone, as was observed in March. In the mixed zone, minor particle retention (60 and 50%) was observed and the remaining particles moved toward to the frontal zone (40 and 50%).

DISCUSSION

The taxonomic contribution of this study is the larval identification of endemic Sciaenidae species in the UGC, a Biosphere Reserve with strong fishery and social controversies (Lercari and Chávez, 2007; Erisman *et al.*, 2010; Bobadilla *et al.*, 2011, Valenzuela-Quiñonez *et al.*, 2014.). The presence of *T. macdonaldi*, *M. megalops*, *Me. nasus* and *C. othonopterus* larvae in the region was corroborated by DNA sequences. In addition, it is the first time that the presence of larvae of the species *I. remifer* is revealed through genetic identification in the UGC. However, because the larvae of the Sciaenidae family are very similar morphologically and the species *I. remifer*, was not previously identified in morphologically analyses, some DNA sequences of *C. othonopterus* were assigned to *Isopisthus remifer*, this was defined as *C. othonopterus-I. remiffer* complex. The definition of a "larval complex" highlights the need to continue with larval identification studies, both morphological and genetic, that allow us to identify other species that inhabit the region.

Our work elaborated on the role of the stratification front ($\Phi = 10\,J/m^3$) on the spawning areas of most of the species analyzed here. Even though the front had

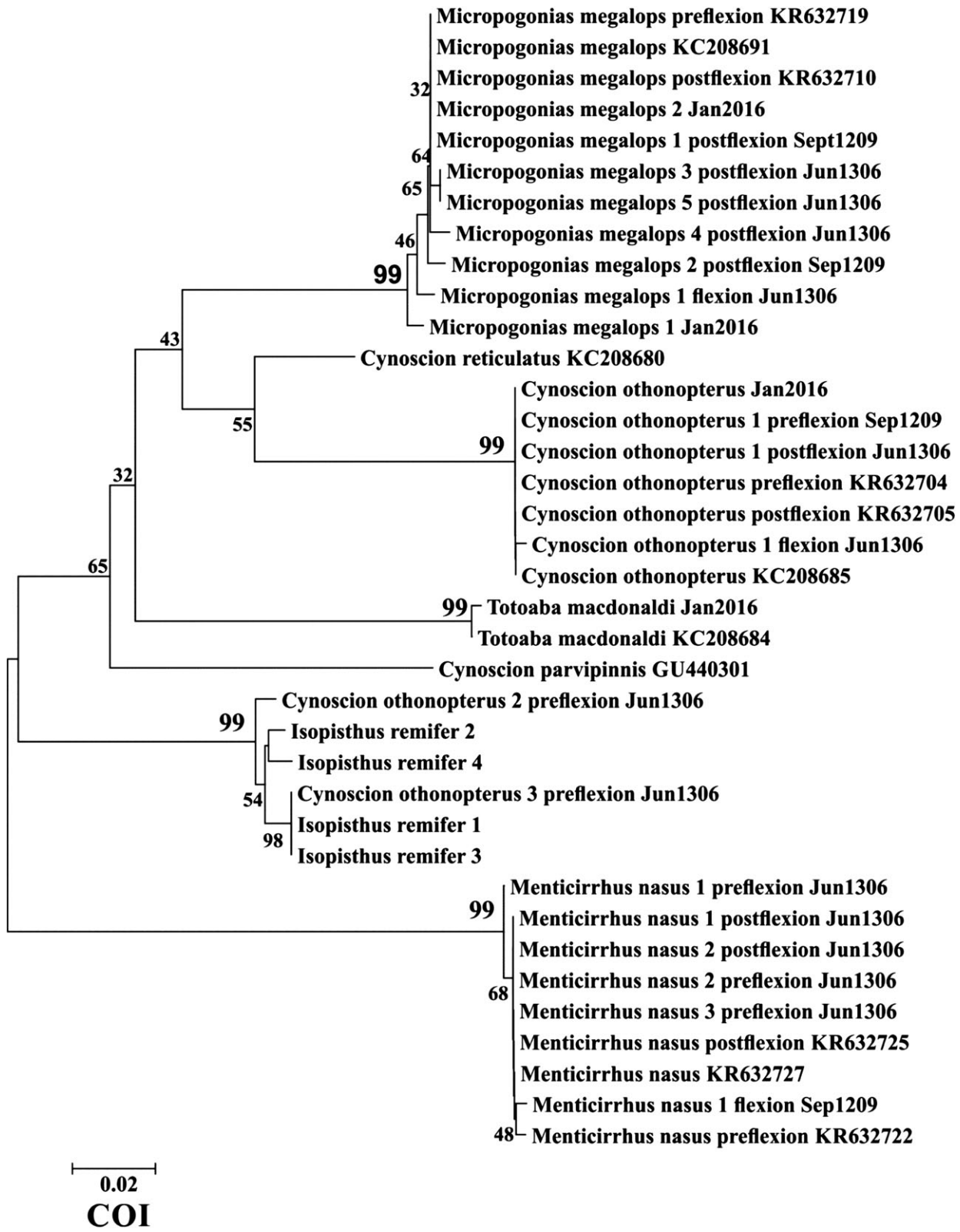


Fig. 6. Nearest neighbor tree based on the variation of the COI gene sequences (486 bp) of mtDNA. The values of the branches indicate the support of the groupings based on 5000 re-sampling replicas. The scale of the bar (0.02) indicates the distance of Kimura's parameter-2.

Table II: Larval abundance (larvae/10 m^2) of Sciaenidae species in the UGC

	Larval fish abundance			
Species	March	June	September	Total
Totoaba macdonaldi	111 [83]			**111 [83]**
Cynoscion othonopterus-Isopisthus remifer complex		1684 [348]	2451 [1881]	**4135 [2229]**
Menticirrhus nasus		253 [51]	322 [245]	**575 [296]**
Micropogonias megalops		2242 [463]	55 [44]	**2297 [507]**
Total	111 [83]	4179 [862]	2828 [2170]	**7118 [3115]**

The numbers in brackets correspond to the sum of the gross value of larvae identified per species.

already been described by authors such as Argote *et al.* (1995) and Lavín *et al.* (1998), its ecological role in the endemism that characterizes the region (Erisman *et al.*, 2012; Valenzuela-Quiñonez *et al.*, 2014) had not been described to date. The stratification front that is located on the southern edge of the UGC, with seasonal variations of its position, is the result of the convergence of high salinity mixed water (37.4 g/kg) from the shallowest northern part (~20 m depth) with less salty stratified water (35.4 g/kg) coming from the adjacent oceanic water (>200 m depth). The hydrographic data analyzed here showed that in June 2013 the front was located further north (~30 m depth) than in March 2011 and September 2011 (~50 m depth). In addition, Argote *et al.* (1995) found that during the summer (September 1986), it was close to ~30 m depth and in winter (December 1986) positioned over ~60 m depth. Although the precise mechanisms that determine the front position are still unknown, it is evident that the seasonality and inter-annual events affect its location.

Even though *T. macdonaldi* larvae were only collected in the northwest area in March, the larval distribution of *M. megalops*, *Me. nasus* and *C. othonopterus- I. remifer* complex was associated with both the front position and season. In June, these larvae were at the front, mainly in the northeastern side of the Gulf; and in September, *Me. nasus* and *C. othonopterus-I. remifer* complex larvae extended their distribution to the north. Independent of the variations among the species spawning periods, sciaenid larvae were not present to the south of the frontal zone, indicating that the stratification front might be functioning like a barrier that prevents the larval advection of coastal demersal species towards the oceanic zone.

The idea that the front constrains dispersal was echoed by the connectivity matrixes. High particle retention was observed in the quadrants that covered the frontal zone, with the highest retention (~>80%) in the eastern side of the UGC. The rest of the particles (~20%) were transported mostly from the eastern to the western side along the frontal zone, which was associated with the pattern of cyclonic circulation during summer in the UGC (Montes *et al.*, 2015). These results correspond with previous studies focused on other zooplankton groups. Soria *et al.* (2012) reported transport of planktonic phases of bivalve mollusc and shrimp from the Sonora coast to the central region of the UGC. It was thus hypothesized that retention processes occurring in the stratification front have a greater influence on the zooplankton organisms than transport by currents. Therefore, the front might be an important nursery area of demersal coastal fish species, and other meroplankton organisms.

The fact that the *T. macdonaldi* larvae were only found in the northwestern side of the UGC, in the shallowest and saltiest region, suggests that the stratification front has no relationship with the distribution of larvae in this species. The spawning area of this species could be associated with other hydrographic factors and the adult habitat. Little is known about this, but their collection in this study corresponded with the spawning season reported by Cisneros-Mata *et al.* (1995), which occurs between February and May, with the highest reproductive activity between March and April. It is clear there is a need to carry out intensive monitoring of the spawning area of the *T. macdonaldi* to understand the spawning response to hydrographic conditions and generate elements that support fisheries management.

The overall results of this study indicate that the stratification front could retain planktonic phases of the Sciaenidae species analyzed here, as well as of other meroplanktonic organisms, preventing their advection outside the UGC. In the case of coastal demersal species, the adults are characterized by their sedentary habits and low mobility, and the front may retain their planktonic stages; this could imply over time, a gradual isolation of the species.

Previous studies of endemism in the Gulf of California reported the presence of more than 90 endemic fish species (Brusca *et al.*, 2005). They speculated that the high endemism might be due to the different ecosystems that occur along of the Gulf and to barriers resulting from permanent oceanographic processes (SEMARNAT, 2004). The hydrographical data

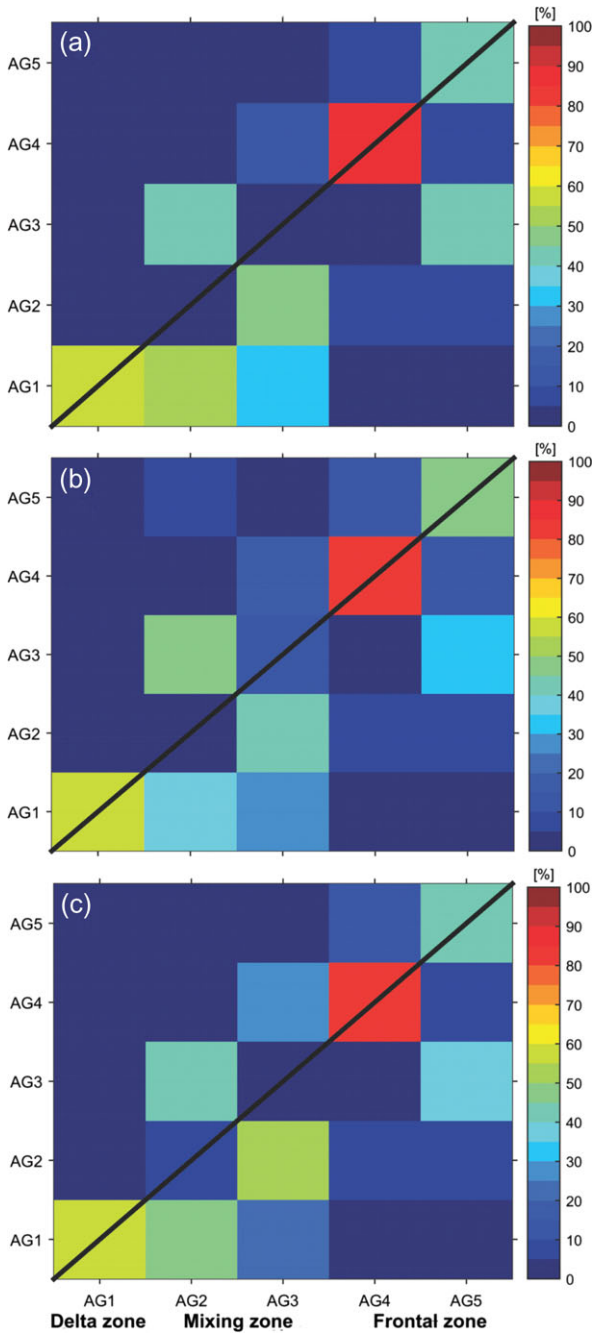


Fig. 7. The connectivity matrices for the months of March (**a**), June (**b**) and September (**c**). The vertical axes correspond to the release areas and the horizontal axes correspond to the arrival areas. The particles in the rows add to 100%. The diagonal lines are visual aids to locate the different elements of the matrices.

in this study show that at least in the UGC, a permanent hydrographical structure, the stratification front, might be a factor that is contributing to the process of endemism in the region. The isolation of populations has been described in ecosystems worldwide in relationship to oceanic fronts that influence processes of geographical isolation and, therefore, lead to genetic isolation (Gilg and Hilbish, 2003, Galarza *et al.*, 2009, Schunter *et al.*, 2011). Therefore, it is important to increase these kinds of studies around permanent hydrographic structures to contribute to the understanding of the changes in the distribution of marine species.

CONCLUSIONS

Genetically based larvae identification of *T. macdonaldi*, *M. megalops* and *Me. nasus* and the definition of the *C. othonopterus-I. remifer* complex allowed definition of their distribution and spawning season. Considering that the *T. macdonaldi* is currently threatened with extinction and the *M. megalops* and *C. othonopterus* support a high fishing effort, the information obtained in this study could be a support to improve the management and conservation of these endemic species in the UGC.

Independent of the variations among the spawning periods of the Sciaenidae species studied here, larvae were not present south of the stratification front, indicating that the front might function as a barrier that prevents the larval advection towards the oceanic zone. Considering that the adults of these species coastal demersal are characterized by sedentary habits and low mobility and larval dispersal seems to be limited, it is probable that genetic isolation resulted in the evolution of endemic species in the UGC.

FUNDING

We thank the project "Ecological Monitoring of the Upper Gulf of California," financed by the David and Lucile Packard Foundation (contract no. 2010–36 137) through the University of Arizona PANGAS. This work also was made possible thanks to the financial support of SEP-CONACyT (contracts 2014-236 864, LSV), and Fronteras de la Ciencia-CONACyT (contracts 2015-2-280, LSV).

ACKNOWLEDGEMENTS

We thank the scientific participants, skipper and crew of the R/V Francisco de Ulloa for their support during the cruise. We thank to Eileen Glazier for the English corrections. Many thanks to Andrew Thompson and other two anonymous reviewers for their comments and contributions to this study.

REFERENCES

Aljanabi, S. M. and Martinez, I. (1997) Universal and rapid salt-extraction of high quality genomic DNA for PCR-based techniques. *Nucleic Acids Res.*, **25**, 4692–4693.

All, J. D. (2006) Colorado river floods, droughts, and shrimp fishing in the Upper Gulf of California, Mexico. *J. Environ. Manage.*, **37**, 111–125.

Anonymous. (2016b) Collateral damage: how illegal trade in Totoaba swim bladders is driving Vaquita to extinction. Environmental Investigation Agency. www.eia-international.org, accessed 14 October 2016.

Argote, M. L., Amador, A., Lavín, M. F. and Hunter, J. R. (1995) Tidal dissipation and stratification in the Gulf of California. *J. Geophys. Res.*, **100**, 103–118.

Bobadilla, M., Álvarez-Borrego, S., Avila-Foucat, S., Lara-Valencia, F. and Espejel, I. (2011) Evolution of environmental policy instruments implemented for the protection of totoaba and the vaquita porpoise in the Upper Gulf of California. *Environ. Sci. Policy*, **14**, 998–1007.

Borguez, R., Vaz, J., Serrao, E. A. and Goncalves, E. J. (2009) Short-term temporal fluctuation of very-near shore larval fish assemblages at the Arrábida Marine Park (Portugal). *J. Coast Res.*, **56**, 376–380.

Brusca, R. C., Findley, L. T., Hastings, P. A., Hendrickx, M. E., Torre, J. and Van der Heiden, M. (2005) Macrofaunal biodiversity in the Gulf of California (Sea of Cortez). In Cartron, J.-L. E. and Ceballos, G. (eds), *Biodiversity, Ecosystems, and Conservation in Northern Mexico*. Oxford University Press, USA, pp. 179–202.

Camacho-Gastélum, R., Díaz-Viloria, N., Sánchez-Velasco, L., Jiménez-Rosenberg, S. P. A. and Perez-Enriquez, R. (2016) Molecular identification and morphological description of *Micropogonias megalops*, *Cynoscion othonopterus*, *C. reticulatus*, and *Menticirrhus nasus* larvae, collected in the upper Gulf of California during summer 2012. *Mitochondrial DNA Part A*, **28**, 416–423.

Carton, J. A. and Giese, B. S. (2008) A reanalysis of ocean climate using Simple Ocean Data Assimilation (SODA). *Mon. Weather Rev.*, **136**, 2999–3017.

Chao, L. N. (1995) Sciaenidae. In Fisher W., Krupp F., Schneider W., Sommer C., Carpenter K. E., Niem V. H. (eds), *Guía FAO para la identificación de especies para los fines de la pesca*. Pacífico Centro-Oriental. Part 2, vol. 3. pp. 1427–1520. FAO, Rome. p. 664.

Cisneros-Mata, M. A., Montemayor-López, G. and Román-Rodríguez, M. J. (1995) Life history and conservation of *Totoaba macdonaldi*. *Conserv. Biol.*, **9**, 806–814.

Cudney-Bueno, R., Lavín, M. F., Marinone, S. G., Raymondi, P. T. and Shaw, W. W. (2009) Rapid effects of marine reserves via larval dispersal. *PLoS One*, **4**, e4140.

DOF. (1993) *Decreto por el que se declara área natural protegida con el carácter de reserva de la biosfera, la región conocida como "Alto Golfo de California y Delta del Río Colorado", ubicada en aguas del Golfo de California en los municipios de Mexicali, Baja California, de Puerto Peñasco y San Luis Río Colorado, Son.* DOF, Mexico.

Díaz-Viloria, N., Sánchez-Velasco, L., Pérez-Enríquez, R. and Jiménez-Rosenberg, S. P. A. (2013) Molecular identification and morphological description of Totoaba (*Totoaba macdonaldi)* and curvina (*Cynoscion reticulatus)* preflexion larvae (Perciformes: Sciaenidae). *Ichthyol. Res.*, **60**, 390–395.

Erisman, B., Aburto-Oropeza, O., González-Abraham, C., Mascareñas-Osorio, I., Moreno-Báez, M. and Hastings, P. A. (2012) Spatial-temporal dynamics of a fish spawning aggregation and its fishery in the Gulf of California. *Sci. Rep.*, **2**, 284.

Erisman, B., Mascarenas, I., Paredes, G., Sadovy de Mitcheson, Y., Aburto-Oropeza, O. and Hastings, P. (2010) Seasonal, annual, and long-term trends in commercial fisheries for aggregating reef fishes in the Gulf of California, Mexico. *Fish. Res.*, **106**, 279–288.

Galarza, J., Carreras-Carbonell, J., Macpherson, E., Pascual, M., Roques, S., Turner, G. and Ciro, R. (2009) The influence of oceanographic fronts and early-life-history traits on connectivity among littoral fish species. *Proc. Natl Acad. Sci. USA*, **106**, 1473–1478.

Gilg, M. R. and Hilbish, T. J. (2003) Patterns of larval dispersal and their effect on the maintenance of a blue mussel hybrid zone in Southwest England. *Evolution*, **57**, 1061–1077.

Hastings, P. A., Findley, L. T. and Heiden, A. V. D. (2010) Fishes of the Gulf of California. In the Gulf of California, biodiversity and conservation. In Brusca, R. C. (ed.), *The University of Arizona Press and the Arizona-Sonora Desert Museum*. Tucson, pp. 96–118.

IOC, SCOR, and IAPSO. (2010) The international thermodynamic equation of seawater—2010: calculations and use of thermodynamic properties. Intergovernmental Oceanographic Commission, Manuals and Guides No.56, UNESCO (English), p. 196.

Ivanova, V. N., Zemlak, S. T., Hanner, H. R. and Hebert, D. N. P. (2007) Universal primer cocktails for fish DNA barcoding. *Mol. Ecol. Notes*, **7**, 544–548.

Kendall, A. W., Jr., Ahlstrom, E. H. and Moser, H. G. (1984) Early life history stages of fishes and their characters, 11-23. In Moser, H. G. (ed), *Ontogeny and Systematic of Fishes*. American Association of Ichthyologists and Herpetologists Special Publication **1**, p. 760.

Kimura, M. (1980) A simple method for estimating evolutionary rate of base substitutions through comparative studies of nucleotide sequences. *J. Mol. Evol.*, **16**, 111–120.

Lavín, M. F., Godínez, V. M. and Alvarez1, L. G. (1998) Inverse-estuarine features of the Upper Gulf of California. *Estuar. Coast. Shelf Sci.*, **47**, 769–795.

Lercari, D. and Chávez, E. A. (2007) Possible causes related to historic stock depletion of the totoaba, *Totoaba macdonaldi* (Perciformes: Sciaenidae), endemic to the Gulf of California. *Fish. Res.*, **86**, 136–142.

Mesinger, F., DiMego, G., Kalnay, E., Mitchell, K., Shafran, P. C., Ebisuzaki, W., Jovic, D., Woollen, J., Rogers, E., Berbery, E. H., Ek, M. B., Fan, Y., Grumbine, R., Higgins, W., Li, H., Lin, Y., Manikin, G., Parrish, D. and Shi, W. (2006) North American regional reanalysis. *Bull. Am. Meteorol. Soc.*, **87**, 343–360. doi:10.1175/BAMS-87-3-343.

Montes, J. M., Lavín, M. F. and Parés-Sierra, A. F. (2015) Seasonal heat and salt balance in the Upper Gulf of California. *J. Coastal. Res.*, **32**, 853–862.

Moser, H. G. (1996) *The Early Stages of Fishes in the California Current Region. CalCOFI Atlas No. 33*. Allen Press, Inc, Luwrence, Kansas, p. 1505.

Norma Oficial Mexicana. (2010) NOM–059–SEMARNAT–2010: Protección ambiental–Especies nativas de México de flora y fauna silvestres–Categorías de riesgo y especificaciones para su inclusión, exclusión o cambio–Lista de especies en riesgo. Diario Oficia, 2010, vol. 30.

Pollnac, R., Christie, P., Cinner, J. E., Dalton, T., Daw, T. M., Forrester, G. E. and McClanahan, T. R. (2010) Marine reserves as

linked social–ecological systems. *Proc. Natl. Acad. Sci. USA*, **107**, 18262–18265.

Proehl, J. A., Lynch, D. R., McGillicuddy, D. J. and Ledwell, J. R. (2005) Modeling turbulent dispersion on the north flank of Georges Bank using Lagrangian particle methods. *Cont. Shelf Res.*, **25**, 875–900.

Rodríguez-Pérez, M., Aurioles-Gamboa, D., Sánchez-Velasco, S., Lavín, M. F. and Newsome, S. D. (2018) Identifying critical habitat of the endangered vaquita (Phocoena sinus) with regional d13 C and d15 N isoscapes of the Upper Gulf of California, Mexico. *Marine Mammal Science*, Vol. 00, No. 00, 2018. Society for Marine Mammalogy. doi:10.1111/mms.12483.

Rodríguez-Quiroz, G., Aragón-Noriega, E. A. and Ortega-Rubio, A. (2010) Artisanal shrimp fishing in the Biosfere Reserve of the Upper Gulf of California. *Crustaceana*, **82**, 1481–1493.

Rojas-Bracho, L., Reeves, R. and Jaramillo-Legorreta, A. (2006) Conservation of the vaquita Phocoena sinus. *Mamm. Rev.*, **36**, 179–216.

Sadovy de Mitcheson, Y. and Erisman, B. E. (2012) The social and economic importance of aggregating species and the biological implications of fishing on spawning aggregations. In Sadovy de Mitcheson, Y. and Colin, P. L. (eds), *Reef Fish Spawning Aggregations: Biology, Research and Management*. Springer, New York, pp. 225–284.

Schunter, C., Carreras-Carbonell, J., Macpherson, E., Tintoré, J., Vidal-Vijande, E., Pascual, A., Guidetti, P. and Pascual, M. (2011) Matching genetics with oceanography: directional gene flow in the Mediterranean fish species. *Mol. Ecol.*, **20**, 5167–5181.

SEMARNAT. (2004) Islands and Protected Areas of the Gulf of California, Mexico. For inscription on the world heritage list. National Cornrnission of Protected Natural Areas. Secretary of Environment and Natural Resources, p. 554.

Simpson, J. H. (1981) The shelf-sea fronts: implications of their existence and behavior. *Philos. Trans. R. Soc. Lond. A*, **302**, 531–546.

Smith, P. E. and Richardson, S. L. (1979) Model techniques for prospection of eggs and larvae of pelagic fish. FAO. Fisheries Technical Documents, N0175, FIR/T175 (Es), p. 107.

Soria, G., Munguía-Vega, A., Marinone, S. G., Moreno-Báez, M., Martínez-Tovar, I. and Cudney-Bueno, R. (2012) Linking bio-oceanography and population genetics to assess larval connectivity. *Mar. Ecol. Prog. Ser.*, **463**, 159–175.

Sánchez-Velasco, L., Lavín, M. F., Jiménez-Rosenberg, S. P. A., Montes, J. M. and Turk-Boyer, P. J. (2012) Larval fish habitats and hydrography in the Biosphere Reserve of the Upper Gulf of California (June 2008). *Cont. Shelf Res.*, **33**, 89–99.

Tamura, K., Stecher, G., Peterson, D., Filipski, A. and Kumar, S. (2013) MEGA6: Molecular Evolutionary Genetics Analysis version 6.0. *Mol. Biol. Evol.*, **30**, 2725–2729.

Valenzuela-Quiñonez, F., Garza, J. C., De-Anda-Montañez, J. A. and Garcia de León, F. J. (2014) Inferring past demographic changes in a critically endangered marine fish after fishery collapse. *ICES J. Mar. Sci.*, **71**, 1619–1628. doi:10.1093/icesjms/fsu058.

Visser, A. W. (2008) Lagrangian modelling of plankton motion: from deceptively simple random walks to Fokker–Planck and back again. *J. Mar. Syst.*, **70**, 287–299.

Ward, R. D., Zemlak, T. S., Innes, B. H., Last, P. R. and Hebert, P. D. N. (2005) DNA barcoding Australia's fish species. *Philos. Trans. R. Soc. Lond. B: Biol. Sci.*, **360**, 1847–1857.