Research Article

Distribution and abundance of *Engraulis ringens* eggs along the north-central Chilean coastline (25.0-31.5ºS) during February 2008 to 2014

Armando Mujica[1], María Luisa Nava[1], Ken Matsuda[2] & Alejandra Vargas[1]
[1]Departamento de Acuicultura, Universidad Católica del Norte, Coquimbo, Chile
[2]Departamento de Matemáticas, Universidad de La Serena, La Serena, Chile
Corresponding author: Armando Mujica (amujica@ucn.cl)

ABSTRACT. In the north-central Chilean coast (25.5-31.5ºS), zooplankton samples were analyzed in 100 oceanographic stations from six oceanographic cruises made in February of 2008, 2009, 2010, 2011, 2013 and 2014. *Engraulis ringens* eggs were separated and counted, thus providing data on distribution, abundance, interannual variation, and relationships with ocean surface temperature and chlorophyll-*a*. The egg distribution was preferentially coastal, with maximum concentrations at stations next to Esmeralda Cove (26ºS) and Chañaral Cove (29ºS). In this time series, which includes cold and warm periods, it was established the relationship of these biological variables with defined ranges of temperature (16,1º-18,0ºC).

Keywords: *Engraulis ringens*, eggs, temperature, interannual distribution, north-central Chile.

Distribución y abundancia de huevos de *Engraulis ringens* en la zona centro-norte de Chile (25,0º-31,5ºS) en febrero 2008-2014

RESUMEN. En la zona centro-norte de Chile (25,0º- 31,5ºS), se analizaron muestras de zooplancton en 100 estaciones oceanográficas de seis cruceros oceanográficos efectuados en febrero de los años 2008, 2009, 2010, 2011, 2013 y 2014. De las muestras se separaron y contabilizaron los huevos de *Engraulis ringens*, para determinar su distribución, abundancia, variación interanual y la relación con la temperatura superficial del mar y concentración de clorofila-*a*. La distribución de los huevos fue preferentemente costera, con máximas concentraciones en estaciones ubicadas próximo a Caleta Esmeralda (26ºS) y Caleta Chañaral (29ºS). En esta serie de tiempo, que incluye periodos fríos y cálidos, se estableció la relación de estas variables biológicas con rangos definidos de temperatura (16,1º-18,0ºC).

Palabras clave: *Engraulis ringens*, huevos, temperatura, distribución interanual, centro-norte de Chile.

INTRODUCTION

Engraulis ringens is an anchovy species with a wide latitudinal distribution across the southwestern Pacific and an important standing in the regional fishing industry (Castro *et al*., 2000; Canales & Leal, 2009; Soto-Mendoza *et al*., 2010). Three *E. ringens* populations have been found, with the distribution of these populations being 1) northern and central Peru, 2) southern Peru, and 3) northern and south-central Chile (Canales & Leal, 2009). Canales & Leal (2009) also found a fishing stock of this species in north-central Chile (25 and 32ºS) that has an independent population unit that recruits, grows, and reproduces in the area.

This species spawns near the ocean surface (0-40 m). Maximum spawning for *E. ringens* primarily occurs along the coastline at a depth of 20 m (Lett *et al*., 2007; Braun *et al*., 2007, 2008, 2009) between August and March, with peaks occurring at the end of the Austral winter (August-September) and during the Austral summer (February-March) (Cubillos *et al*., 1999; Perea *et al*., 2011; Hernández-Santoro *et al*., 2013). The spawning, abundance, and egg distribution in plankton of *E. ringens* have been related to a number of environmental variables, including temperature, salinity, chlorophyll-*a* levels, and upwelling (Escribano *et al*., 1996; Braun *et al*., 2007; Tarifeño *et al*., 2008; Soto-Mendoza *et al*., 2010; Claramunt *et al*., 2012).

Corresponding editor: Guido Plaza

Nevertheless, spawning has not been uniquely associated with any of these or other environmental variables, which is a product of this species wide latitudinal distribution, broad spawning period, and the existence of discrete populations within the distribution area. These variances, in turn, are the result of the diverse range of environmental variables existing within the geographical areas of this species.

Among the determined environmental variables, the temperature at which spawning occurs has evidenced recurring patterns, with spawning in northern Chile occurring between 15 and 18°C and in south-central Chile, between 12 and 15°C (Tarifeño *et al*., 2008). Tarifeño *et al*. (2008) additionally linked spawning with the lower temperatures that occur in upwelling zones, which are also nutrient rich.

By evaluating the second spawning (February) of *E. ringens* over a number of years, the present study associated *E. ringens* egg abundances and distributions with ocean surface temperature and chlorophyll-*a* concentration.

MATERIALS AND METHODS

Zooplankton samples were taken onboard the research vessel B/C Abate Molina of the Instituto de Fomento Pesquero (IFOP) in February 2008, 2009, 2010, 2011, 2013, and 2014 (Fishery Improvement Project [FIP]: Evaluación hidroacústica del reclutamiento de anchoveta entre la III y IV Regiones). Samples were collected at 80 oceanographic stations located 1, 5, 10 and 20 nm (nautical miles) from the coast and at an additional 20 oceanographic stations located 1 nm from the coast. All stations were distributed within 20 transects perpendicular to the coast between Paposo (25.0ºS) and Puerto Oscuro (31.5ºS), Chile (Fig. 1). The stations were always sampled from north to south during the first and last days of February each year (25 consecutive days).

Bongo nets (59 cm diameter, 300 µm mesh) equipped with flowmeters were used to collect zooplankton between the surface and a depth of 70 m, or 10 m from the bottom at sites with a lesser depth. The samples were preserved with a formalin solution in 5% seawater, from which *E. ringens* eggs were separated and quantified (number of eggs 100 m^{-3}). The abundance, dominance, and frequency of *E. ringens* eggs were determined overall and in regards to site distance from the coast (1, 5, 10, and 20 nm).

Ocean surface temperature and integrated chlorophyll-*a* (chl-*a*) levels (0-100 m), obtained from Castillo *et al*. (2009a, 2009b, 2010, 2012, 2013) and Leiva *et al*., (2014), were related to *E. ringens* egg abundances, distributions, and frequencies through *Q* coefficient analysis (Van der Lingen *et al*., 2001; Bernal *et al*., 2007; Claramunt *et al*., 2012). For this, five temperature ranges (≤14; 14.1-16; 16.1-18; 18.1-20; and >20°C) and six chl-*a* ranges (≤20; 20.1-40; 40.1-60; 60.1-80; 80.1-100; and > 100 mg m^{-2}) were established.

$$Q = \frac{(n°h_r * 100)/n°h_t}{(n°est_r * 100)/n°est_t}$$

where $n°h_r$: number of eggs in the ***r*** range (temperature or chlorophyll-*a*), $n°h_t$: total number of eggs, $n°est_r$: number of sampling stations in the *r* range, and $n°est_t$: total number of sampling stations.

To determine statistically significant differences of *Q* within the distinct temperature ranges, chl-*a* ranges, and sampling years, the normal distribution of the dependent variable (*Q)* was confirmed by the Kolmogorov-Smirnov test while homoscedasticity was verified by the Lavene test ($P < 0.05$).

Following this, randomized blocks of the sampling years (2008, 2009, 2010, 2011, 2013, and 2014) were tested with an analysis of variance, using years, temperature range, chl-*a*, and the variable response to *Q* as factors. Significantly different variables were later analyzed by a Tukey test.

Since the maximum values of the *Q* coefficient were found within different chl-*a* range in different sampling years, Bootstrap analysis was used to confirm randomization and to verify if the maximum *Q* value was significant for each year.

RESULTS

The greatest total abundance of *E. ringens* eggs (>150,000 eggs 100 m^{-3}) was found during February 2010, reaching an order of magnitude greater than the other years (Table 1). Egg frequency was within 15 and 32% at each sampling station. Egg distributions showed similar patterns for all years, with preferential distribution towards coastal sites. The dominance at stations 1 nm from the coast was always greater than 94%.

Engraulis ringens eggs were collected along the coastline from the extreme northern zone of the study area (25°S) to the Bay of Tongoy (30°10’S). Only in February 2008 were eggs collected south of the Bay of Tongoy, and the distribution of eggs at these sites was notably different than at more northern sites, evidencing the minimum recorded values of abundance and frequency found over the years studied (Fig. 2). The zone between Caldera and Chañaral Cove (27-29ºS) generally showed low egg abundances, and in February 2008, no *E. ringens* eggs were found. However, in February 2010, 2013, and 2014, eggs were found in

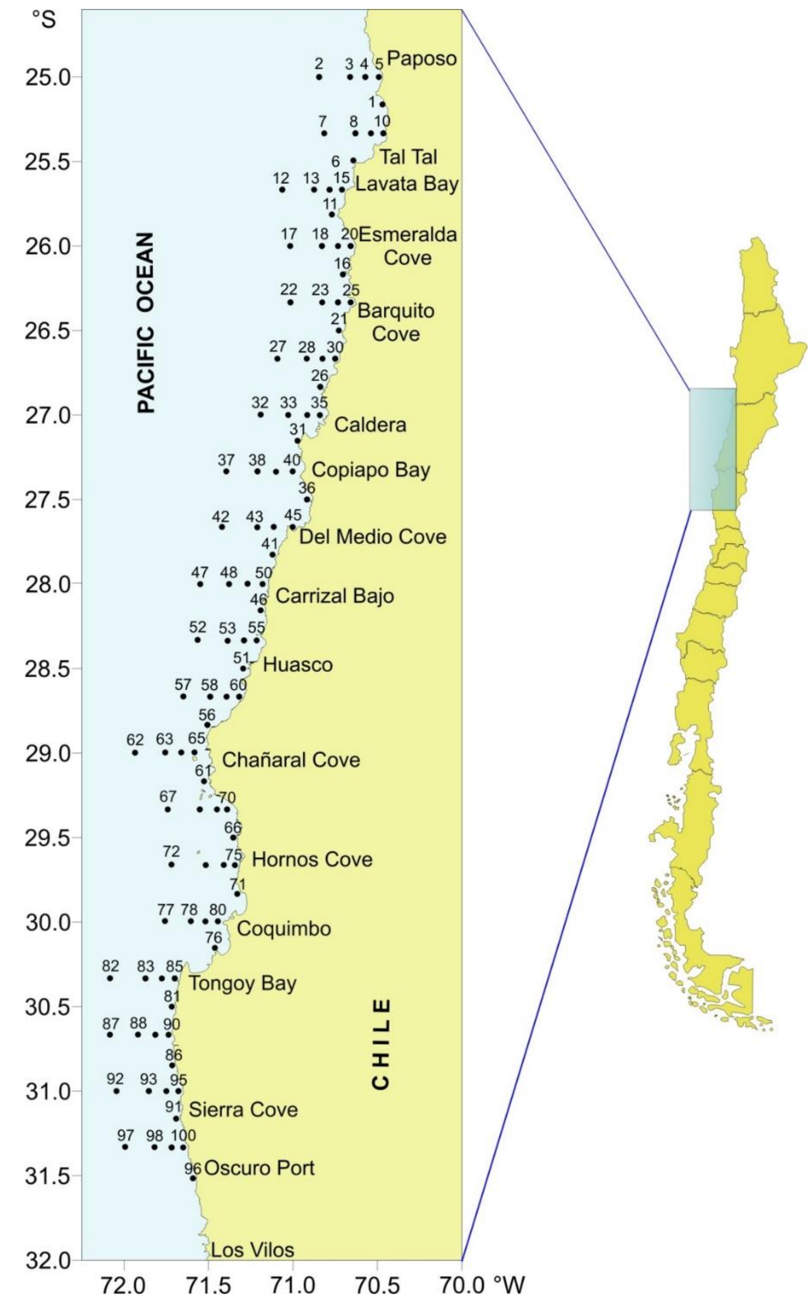

Figure 1. Sampling stations location. Samples were taken during February 2008, 2009, 2010, 2011, 2013, and 2014.

these zone even at the most coastally distant stations (Fig. 2).

The maximum egg abundances were always recorded at stations 25 and 70, both of which were coastal and located south of Esmeralda Cove (26ºS) and Chañaral Cove (29ºS), respectively. Only in February 2013 were eggs not collected from station 70 (Figs. 1-2).

The surface temperature at the different stations was between 13 and 24°C, with a clear north-to-south gradient and extreme values in February 2013 (Castillo *et al*., 2009a, 2009b, 2010, 2012, 2013; Leiva *et al*., 2014) (Fig. 3). Regarding integrated chl-*a*, maximum values (>400 mg m^{-2}) were recorded in February 2013. In the first three years of study (2008, 2009, and 2010), chl-*a* values were between 20 and 100 mg m^{-2} (Fig. 4). In general, the lowest ocean surface temperatures and highest integrated chl-*a* were related to a greater upwelling index (Castillo *et al*., 2009a, 2009b, 2010, 2012, 2013; Leiva *et al*., 2014).

Table 1. *E. ringens* egg abundance (number of eggs 100 m^{-3}), dominance (%), and frequency (%) at the sampling stations, grouped by distance from the coast.

		Feb-08	Feb-09	Feb-10	Feb-11	Feb-13	Feb-14
1 nm	Abundance	19,363	35,014	164,038	66,791	79,04	55,622
	Dominance	99.79	97.79	95.62	94.35	94.15	94.76
	Frequency	30.00	45.00	32.5	43.59	43.59	34.21
5 nm	Abundance	41	551	402	3,428	1,333	2,414
	Dominance	0.21	1.54	0.23	4.84	1.59	4.11
	Frequency	15.00	20.00	15.00	42.11	31.58	20.00
10 nm	Abundance	0	238	2,777	568	3,457	488
	Dominance	0.00	0.66	1.62	0.80	4.12	0.83
	Frequency	0.00	10.00	25.00	20.00	11.76	15.00
20 nm	Abundance	0	4	4,331	5	122	17
	Dominance	0.00	0.01	2.52	0.01	0.15	0.03
	Frequency	0.00	5.26	35.00	5.00	26.67	5.26
Total	Abundance	19,405	35,806	171,548	70,791	83,951	58,701
	Frequency	15.00	25.25	28.00	30.61	32.22	23.71

The *Q* coefficient was established between temperature ranges and the presence of *E. ringens* eggs over the years at all stations and at stations 1 nm from the coast. Values greater than 1 (greater affinity between variables) were found for temperature ranges between 16 and 18°C, with the exception of February 2013, where a greater association was established with a range of 18-20°C (Table 2).

On the other hand, the relationship established by the *Q* coefficient between the presence of eggs and the ranges of integrated chl-*a* levels (0-100 m), for all stations and for those located 1 nm from the coast, generated values higher than those established for the temperature ranges. This was especially notable for 2009 and 2010 (60-80 mg m^{-2}). In these years, the majority of the eggs (65 and 44%) were found at stations 4 and 3, respectively (Table 3).

When using randomized blocks in the analysis of variance, statistically significant differences were established between the values of the *Q* coefficient and the temperature ranges for all years and in an integrated time-series, results which are contrary to that established for the ranges of chl-*a* (Table 4). After applying the Tukey test to the integrated time-series, it was possible to establish that the greatest affinities occurred between the temperature ranges 16-18°C and 18-20°C (Table 5). In contrast, the chl-*a* ranges did not evidence significant differences, with the highest affinities found for all ranges ≥3 (>40 mg m^{-2}).

Given that the maximum values of the *Q* coefficient were found within different chl-*a* range over the studied years, a Bootstrap analysis was used to verify that the maximum *Q* value was significant for each year, in addition to confirming randomization. For this, 1000 bootstrap replications were performed, from which the maximum *Q* values were determined for each study year and for both the range of total chl-*a* for all stations and for stations 1 nm from the coast. This also indicated the respective frequency (F), probability (P), and 5% confidence interval for the estimated proportion of *Q* values (Tables 6-7).

The occurrence probabilities for each *Q* value were found greater than 5% for all stations and for those located 1 nm from the coast. The maximum *Q* of the information for each sampling year was estimated by 1000 bootstrap replications, and, for 2008, 694 were found to be greater than or equal to 9,801. This result indicates that the occurrence probability of this value is 694/1000, or, in other words, that 69.4% of the iterations of the *Q* values were equal to 9,801. These calculations determined that the *Q* value was significant for 2008 and that this depended on the distribution of environmental and biological variables. Similar observations were made for the remaining sample years (Tables 6-7).

DISCUSSION

The sampling period (February) of *E. ringens* coincided with the second spawning of this species (Cubillos *et al*., 1999; Perea *et al*., 2011; Hernández-Santoro *et al*., 2013). Due to this, the obtained data regarding distribution, abundance, and interannual variation of the number of eggs collected are representative of the second spawning period.

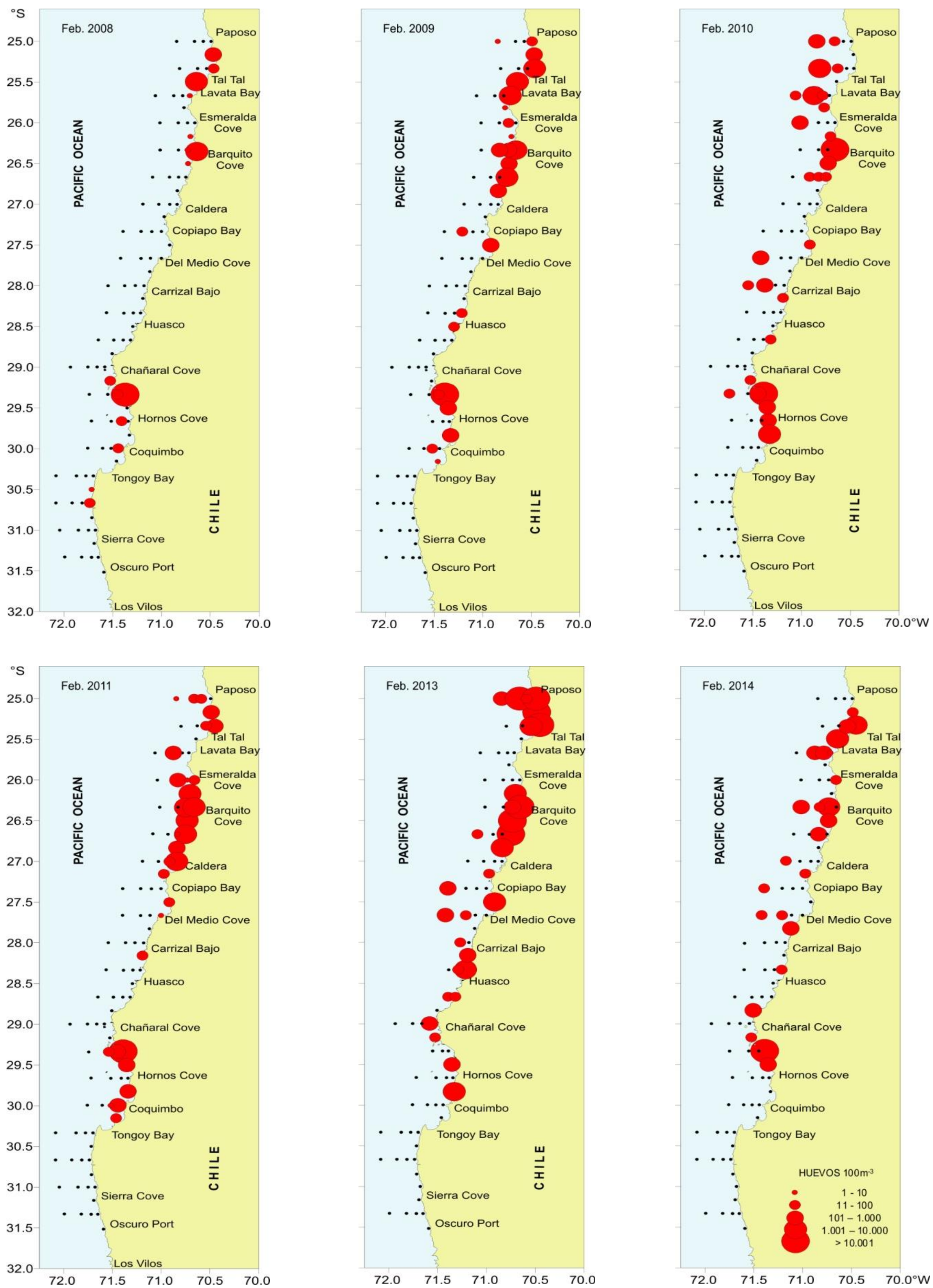

Figure 2. *Engraulis ringens* egg distribution and abundance within the sampling area. Samples were taken during February 2008, 2009, 2010, 2011, 2013, and 2014.

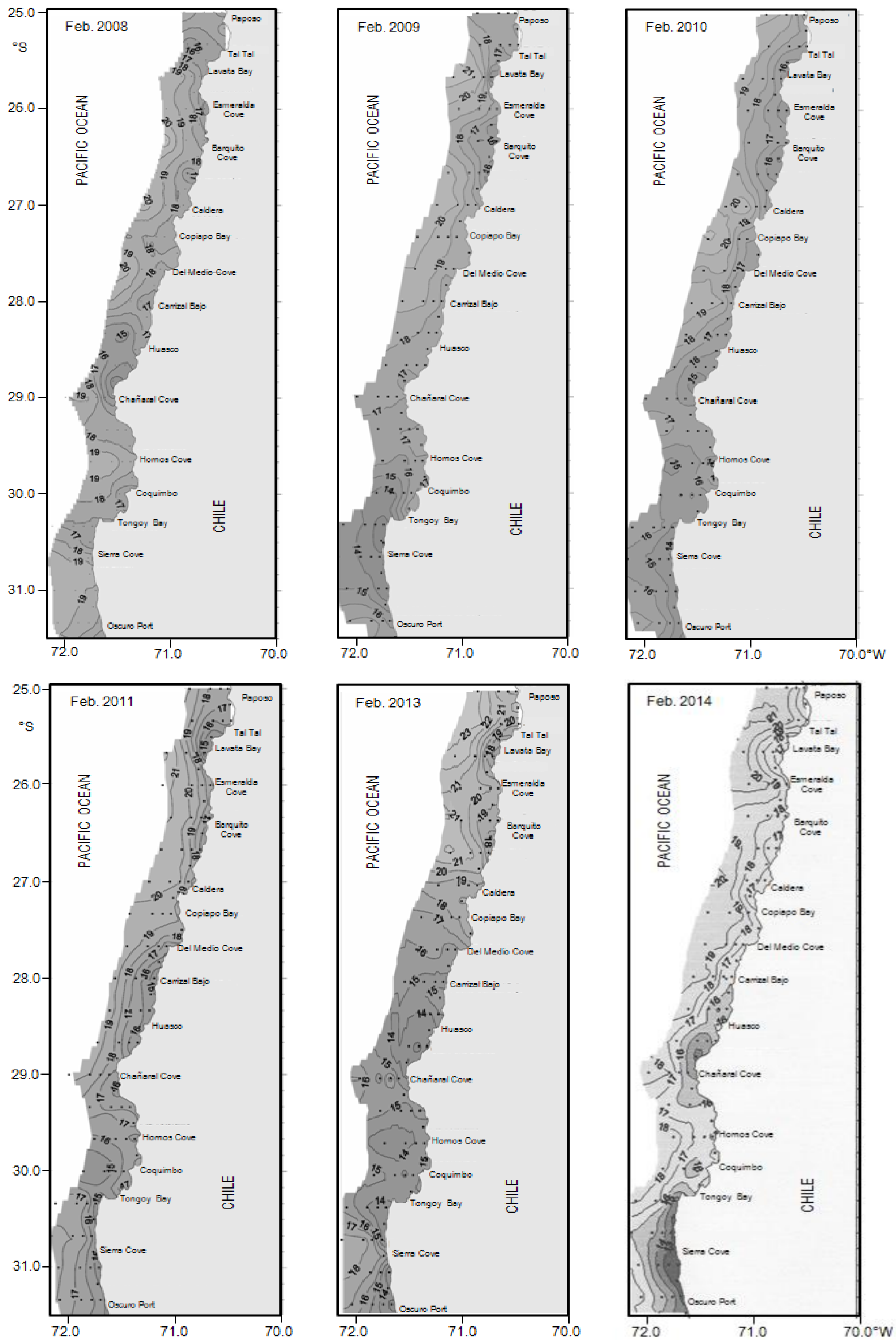

Figure 3. Ocean surface temperature (°C) of the sampling area. Samples were taken during February 2008, 2009, 2010, 2011, 2013, and 2014.

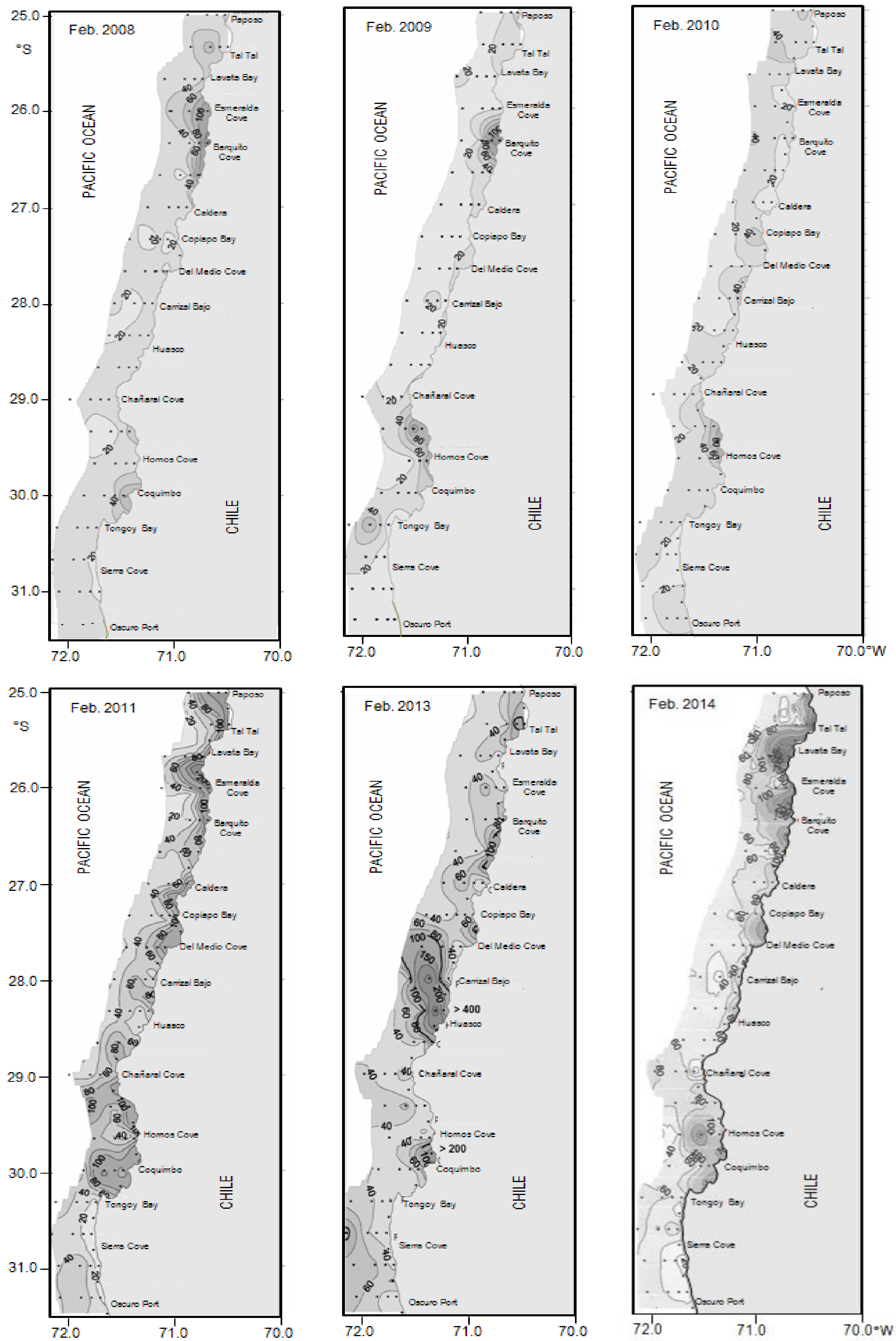

Figure 4. Chlorophyll-*a* (mg m^{-2}) integrated values (0-100 m) within the sampling zone. Samples were taken during February 2008, 2009, 2010, 2011, 2013, and 2014.

Table 2. *Q* coefficient values presented for all sampling stations and those 1 nm from the coast, grouped by temperature range (°C).

	T range (°C)	Feb-08	Feb-09	Feb-10	Feb-11	Feb-13	Feb-14
All stations	≤14.0	0.344	0.000	0.000	0.000	0.001	0.074
	14.1 - 16.0	0.074	**1.227**	0.084	0.036	0.105	0.009
	16.1 - 18.0	**2.008**	**1.796**	**2.163**	**2.174**	0.514	**2.514**
	18.1 - 20.0	0.009	0.044	0.213	0.638	**3.338**	0.212
	>20.0	0.000	0.000	0.139	0.001	0.386	0.000
Stations at 1 nm	≤14.0	0.138	0.000	0.000	0.000	0.000	0.048
	14.1 - 16.0	0.077	**1.056**	0.068	0.024	0.089	0.007
	16.1 - 18.0	**1.459**	**1.447**	**2.041**	**2.128**	0.646	**2.002**
	18.1 - 20.0	0.013	0.065	0.000	0.952	**2.891**	0.390
	>20.0	0.000	0.000	0.000	0.000	0.569	0.000

Table 3. *Q* coefficient values presented for all sampling stations and those 1 nm from the coast, grouped by integrated chlorophyll-*a* levels (0-100 m; mg m^{-2}).

	Chlorophyll-*a* range	Feb-08	Feb-09	Feb-10	Feb-11	Feb-13	Feb-14
All stations	≤20.0	0.021	0.059	0.022	0.000	0.000	0.000
	20.1 - 40.0	0.024	0.868	0.921	0.000	0.087	0.000
	40.1 - 60.0	**9.801**	0.137	0.246	0.241	0.130	0.040
	60.1 - 80.0	0.020	**16.092**	**14.637**	0.892	**1.623**	0.030
	80.1 - 100	0.000	0.369	0.000	**1.210**	**8.649**	**5.342**
	>100	**2.447**	**3.238**	0.000	**4.197**	**1.048**	0.928
Stations at 1 nm	≤20.0	0.019	0.070	0.019	0.000	0.000	0.000
	20.1 - 40.0	0.029	0.875	0.931	0.000	0.001	0.000
	40.1 - 60.0	**5.050**	0.076	0.000	0.043	0.202	0.113
	60.1 - 80.0	0.016	**8.865**	**6.123**	0.821	**2.608**	0.029
	80.1 - 100	0.000	0.004	0.000	0.908	**5.572**	**3.832**
	> 100	0.981	**1.338**	0.000	**2.357**	0.784	0.402

Table 4. Randomized blocks in analysis of variance applied to the *Q* coefficient values for temperature and chlorophyll-*a*.

	Variation source	*SS*	GL	MC	F	*P*
Temperature	Years	7.584	5	1.517	0.956	0.448
	Range	14.651	4	3.663	2.309	**0.052**
	Error	172.884	109	1.586		
Chlorophyll-*a*	Years	9.419	5	1.884	0.517	0.763
	Range	20.388	5	4.078	1.119	**0.354**
	Error	484.778	133	3.645		

The primarily coastal distribution of *E. ringens* eggs found by this study is consistent with Braun *et al*. (2007, 2008, 2009) and Soto-Mendoza *et al*. (2010). These authors reported the presence of *E. ringens* eggs near the ocean surface along the coastal zone within the entire distribution area of this species, with maximum presence found in northern Chile and southern Peru. In turn, the similar interannual distribution pattern of eggs corresponded with those areas showing greater adult abundances of this species, with two coastal sites (25-27º40' and 29-30º10'S) consistently found as focal points for *E. ringens* (Castillo *et al*., 2009a, 2009b, 2010, 2012, 2013; Leiva *et al*., 2014).

The highest concentration of eggs was found in February 2010, coinciding with the lowest total abundance acoustically detected for this species, although the total biomass was the highest found over the sampling years (Castillo *et al*., 2010). A similar

Table 5. Affinity and significance (Tukey test, $P < 0.05$) between the temperature ranges of the Q coefficient values in the time-integrated series.

	Range	n	Subset	*P*
Temperature	1	24	0.00991	0.0492
	5	24	0.46533	
	2	24	0.71292	
	3	24	**0.91992**	
	4	24	**0.97329**	

tendency was found in February 2013 (Castillo *et al.*, 2013), thus involving the greatest proportion of adults in the biomass, which would explain the abundance of eggs. Regarding this, the distributions and abundances of the eggs collected over the different years of study were related to the highest proportions of adults, as acoustically detected over the same sampling periods (Castillo *et al.*, 2009a, 2009b, 2010, 2012, 2013, Leiva *et al.*, 2014). Moreover, the adult distribution of this species is in line with the sites that had the highest egg concentrations; the coast south of Esmeralda Cove (26ºS) and Chañaral Cove (29º40'S), corresponding to stations 25 and 70, respectively. The exception to this tendency, in February 2013 at station 70, could be a result of the predominance of juvenile recruits acoustically detected during this year (Castillo *et al.*, 2013).

Table 6. Bootstrap analysis between the Q coefficient values of chlorophyll-*a* range in the time-integrated series for all stations.

Chlorophyll-*a* range for all stations						
	Max *Q*	F	*P*	Iterations	Lower limit	Upper limit
Feb-08	9.801	694	0.694	1000	693.97	694.03
Feb-09	16.092	665	0.665	1000	664.97	665.03
Feb-10	14.637	686	0.686	1000	685.97	686.03
Feb-11	4.197	680	0.680	1000	679.97	680.03
Feb-13	8.649	630	0.630	1000	629.97	630.03
Feb-14	5.342	656	0.656	1000	655.97	656.03

Table 7. Bootstrap analysis between Q coefficient values of the chlorophyll-*a* range in the time-integrated series for stations at 1 nm.

	Max *Q*	F	*P*	Iterations	Lower limit	Upper limit
Feb-08	5.05	671	0.671	1000	670.97	671.03
Feb-09	8.865	677	0.677	1000	676.97	677.03
Feb-10	6.123	655	0.655	1000	654.97	655.03
Feb-11	2.357	646	0.646	1000	645.97	646.03
Feb-13	5.572	641	0.641	1000	640.97	641.03
Feb-14	3.832	666	0.666	1000	665.97	666.03

A number of authors have related spawning and the abundance of *E. ringens* eggs with environmental variables and oceanographic events. Escribano *et al.* (1996) found that in northern Chile, *E. ringens* spawning is associated with lower seawater temperatures related to upwelling events. In contrast, Claramunt *et al.* (2012) indicates that temperature is not a relevant variable for determining the geographic position of spawning sites for *E. ringens* and that high chlorophyll-*a* concentration is the variable that determines changes in the spawning site. On the other hand, in the zone between Constitución (35º20'S) and Talcahuano (36º42'), Soto-Mendoza *et al.* (2010) did not find a relationship between salinity and the abundance of *E. ringens* eggs and larvae, observing instead that greater egg concentrations can be found outside of the continental water plumes.

Within the period assessed by the present study, Castillo *et al.* (2013) detected colder (February 2008, 2009, and 2011) and warmer (February 2010 and 2013) years, which can be related to the interannual variation of the Humboldt current (Escribano *et al.*, 2002). Associated with this, greater abundances of *E. ringens* eggs were found during the warmer years within the evaluated time period.

The integrated chl-*a* value generally followed the previously indicated interannual ocean surface temperature variations. However, some latitudinal variations were found for chl-*a* level that did not align with egg distributions and abundances in the evaluated years. Regarding this, Claramunt *et al*. (2012) found wide variations between temperature and chl-*a* ranges and the spawning sites of this species, in relation to both the zones analyzed (northern and southern Chile) and whether the studied year evidenced the El Niño phenomenon or not.

The wide latitudinal distribution of this species along the coasts of the south Pacific (Castro *et al*., 2000; Canales & Leal, 2009; Soto-Mendoza *et al*., 2010; Medina *et al*., 2015), the two reported spawning periods (August-September and February-March) (Perea *et al*., 2011), and the association of spawning with different temperatures (Escribano *et al*., 1996; Claramut *et al*., 2007; Soto-Mendoza *et al*., 2010) indicate that different fractions of the *E. ringens* stock spawn under different environmental conditions. During the assessed period, which covered a wide area of distribution (25-31ºS) in February (summer spawning) of consecutive years, use of the *Q* coefficient established that the abundance of eggs was related to ocean surface temperature (16.1-18.0ºC in February 2008, 2009, 2010, 2011, and 2014), as well as to the total number of stations and those located 1 nm from the coast. In February 2013, abundance was related to a temperature range of 18.1-20.0ºC.

The relationship between egg abundances and the indicated temperature ranges, as supported by the *Q* coefficient and statistical analyses, is consistent with previous studies, where during the same study period Castillo *et al*. (2013) identified hotter and colder years.

ACKNOWLEDGEMENTS

This study was funded by projects awarded by the Fondo de Investigación Pesquera (FIP) 2007-03; 2008-02; 2009-03; 2010-03; 2012-13; and 2013-04 (Evaluación hidroacústica del reclutamiento de anchoveta entre la III y IV Regiones). The authors would also like to thank the support provided by the Instituto de Fomento Pesquero (IFOP) that, in partnership with the Universidad Católica del Norte, permitted us to obtain the information used as a starting point for this study. We would also like to thank the crew of the IFOP research vessel B/C Abate Molina and the personnel who aided in sample collection.

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Received: 3 March 2014; Accepted: 26 October 2015

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