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MICROBIOLOGY

Effects of temperature and salinity on the development and survival of the embryos and zoeae I from the southern surf crab *Ovalipes trimaculatus* (Brachyura: Portunidae)

ANTONELA MARTELLI & PEDRO J. BARÓN

Abstract: *Ovalipes trimaculatus* is a highly valued fisheries resource with high potential for aquaculture production. Still, there is need for experimental information to sustain efficient husbandry practices**.** In this work we analyze the combined effects of different thermo-haline conditions on the length of development and survival of embryos (6, 10, 13, 15, 18, 22, 24 ^{0}C x 30, 33 ‰; 13 ^{0}C x 26, 30, 33, 37 ‰) and zoeae I (13, 18, 22 ^{0}C x 30, 33 ‰; 13 ^{0}C x 26, 30, 33, 37 ‰) from individuals sampled in the Atlantic coast of Patagonian. Among the most relevant results, it was found that the mean length of embryogenesis decreased from 63 to 19 days with increasing temperatures, but was not affected by seawater salinity. Mean embryonic survival was significantly lower at the combination of the highest temperature and salinity tested. Also, it differed between salinity conditions. Both at 30 and 33‰, the length of the Zoea I stage significantly varied between thermal treatments, being significantly longer at 13^{0}C. No zoeae I reared at 13 ^{0}C survived at 37‰ and mean survival at 26‰ halved that of 30-33‰. Results obtained reduce aquaculture production costs.

Key words: Temperature, environmental conditions, hatchery, swimming crab.

INTRODUCTION

Temperature and salinity are some of the main modulators of development, growth and survival of different ontogenetic stages of marine invertebrates, including the crustaceans (Anger 2001, Dohle et al. 2004, Scholtz & Wolff 2013). Although temperature produces the most outstanding effects on the length of embryonic and larval development, both variables act together regulating different biochemical metabolic processes and determining growth and survival (Giménez & Anger 2001, Hamasaki 2003, Bas et al. 2008, Sainz-Hernández et al. 2016). Therefore, to fully understand their synergic effects, experimental work must be conducted by testing the effects of both variables simultaneously (Papadopoulos et al. 2006, Fowler et al. 2011).

During larval life, the range of tolerance of marine crustaceans to those, and other environmental variables such PCO_2 in acidified seawater (Schiffer et al. 2014) or the concentration of marine pollutants (Lezcano et al. 2015), fluctuates depending on the species' life history and the association of the different ontogenetic stages to particular habitats (Choy 1991, Charmantier 1998, Tankersley & Forward 2007). The length of different larval growth stages decreases as temperature approaches to an optimum value (Hamasaki 2003, Luppi et al. 2003, Gong et al. 2015, Ikhwanuddin et al. 2016), and as it shortens, the accumulated biomass in each instar diminishes (Anger 2001). Water

salinity affects larval development rates, survival and size (Luppi et al. 2003, Castejón et al. 2015, Ikhwanuddin et al. 2016) while incorporation or elimination of water and salts compensate for osmotic gradient between the body tissues and the surrounding environment (Bas & Spivak 2000).

Manipulation of environmental parameters is a common practice in hatchery and nursery facilities, directed to accelerate marine crustacean larval production and increase survival and fitness (Allan & Fielder 2004, Costejón et al. 2015, Gong et al. 2015, Rahman et al. 2017). However, previous to the design of this technique it is necessary to count with information on thermo-haline tolerance ranges of embryos and larvae, and on the optimum thermo-haline combinations within these resulting in the highest growth and survival (Nurdiani & Zeng 2007). Understanding of the effects of environmental variables on the length and success of embryonic and larval development of marine organisms, on the other hand, can be used in predictive models of larval recruitment success useful for fisheries management and conservation (Barón 2002, Cinti 2002, Crespi-Abril & Barón 2012).

Ovalipes trimaculatus is a portunid crab with a wide geographical distribution along the Atlantic coast of South America, spanning from Southern Brazil (Severino-Rodrigues et al. 2018) to San Jorge Gulf (Patagonia) (Vinuesa 2005). Also, according to presently accepted interpretation of its taxonomic status, the species is distributed on the margins of the Southeast Atlantic, Southeast Pacific, Indian Oceans (Schoeman & Cockcroft 1993), and probably on the South Pacific Ocean (DiSalvo et al. 1988, Boyko & Liguori 2014). Along the Atlantic coast of South America, the species is adapted to average summer and winter seawater temperatures ranging within 12.9-26.9 °C and 5.9-20.9 °C respectively (Baldoni et al. 2015). Although, it is found in environments with typical oceanic water salinities (e.g., ~32.9-33.8 psu in Nuevo Gulf, Rivas & Ripa 1993, average 32.8 psu in coastal waters off Argentina, Baldoni et al. 2015), and it is considered a stenohaline species (Du Preez & McLachlan 1984), adults and megalopae have been observed in Valdivia River mouth (Los Molinos, Chile; 39° 51' S, 73° 24' W) (Pardo et al. 2020) where the confluence of riverine and oceanic waters generate an estuarine condition fluctuating between 27 and 35 psu depending on depth and tidal condition (Garcés-Vargas et al. 2013). Although no study is still available on the osmoregulatory physiology of the species, it can be expected that it acts as an osmoconformer at such salinity range, as its congener *Ovalipes ocellatus* (Birchard et al. 1982). Furthermore, it has been demonstrated that juveniles show higher growth in captivity at salinities slightly lower than those typical from marine environments (Martelli et al. 2019).

While fisheries statistics poorly reflect its annual landings, recent studies show that in northern Patagonia captures with similar proportions of males and females have increased during the last years (de la Barra 2018), posing a risk for the reproductive potential of its populations. Furthermore, due to the high meat quality, and increasing demand of its products (Dima et al. 2012, 2014, 2016, Santos et al. 2012), it is expected that fishing pressure on this resource will intensify. Albeit Argentina has not yet developed a dynamic marine aquaculture industry, several recommendations have been pointed out for its consolidation, including the promotion of strategic differentiation of its products, targeting non-saturated markets with still low tariff barriers for potential competitors, and the identification of local culture species with unsatisfied demand or having the potential to supply niche markets (Burguener & Barón 2016). On these regards, not

only *O. trimaculatus* stands out as an excellent option for the development of the local aquaculture enterprises, but also its culture might compensate market demands, relieving the extractive pressure on its populations.

The embryonic development of *O. trimaculatus* has been studied at a single combination of temperature and salinity (Martelli et al. 2016); still, is necessary to conduct experimental work to determine the optimal combined thermo-haline conditions leading to maximum survival and highest development rate. On the other hand, even when information is available on the survival of *O. trimaculatus* zoeae I from the Indian Ocean (Algoa Bay, South Africa) exposed for a brief period (≤ 24 h) to several thermo-haline conditions (Du Preez & McLachlan 1984), neither this information fulfills the requirements for aquaculture practice design and larval recruitment modelling, nor its extrapolation to individuals from populations on the coasts of South America can be considered reliable. Based on these considerations, the objective of this study is to evaluate the length and success of embryonic and zoeae I development at different combinations of temperature and salinity, aiming to generate key information for the development of hatchery protocols and use in models of early-life-stage population dynamics. Also, it is expected that the results will be useful to complement the available information on the species life cycle.

MATERIALS AND METHODS

Sample collection and conditioning

A total of 121 *O. trimaculatus* females holding masses of newly fecundated eggs in their pleopods were hand-collected by SCUBA diving on shallow (2-9-m deep) subtidal sand bottoms of Nuevo Gulf (42º25´S, 64° 07´W, Argentina) during the reproductive season (October-December) of 2015. Specimens were transported during ~30-m trips from the beach to the Experimental Marine Aquarium of the National Patagonian Sci-Tech Center (CCT CONICET-CENPAT) within plastic refrigerators, cushioned with fresh macroalgae to maintain temperature and humidity. At the time of arrival to the aquarium, specimens were acclimated in lidded plastic tanks (85 × 85 × 60 cm) containing filtered seawater at 13° ± 1°C and 33 ± 1‰, simulating the average SST and salinity conditions through the reproductive season in Nuevo Gulf (Rivas & Ripa 1989, Dellatorre et al. 2012), and were provided with continuous aeration, a layer of macroalgae to use as refuge, and a 12:12 h photoperiod simulating the natural light cycle at the experimental period. Just after arrival to the aquarium, embryo samples were taken from the ovigerous mass of each female and staged under dissecting microscope following the scale of embryonic development developed by Martelli et al. (2016). Ovigerous females carrying embryos at intermediate or advanced stages of development were returned to the wild.

Experimental design

Two sets of experiments were planned to solve limitations in aquaria time and space availability. The first tested the effects of combinations of seven temperatures (6, 10, 13, 15, 18, 22, 24 ºC) and two salinities (30 and 33‰) on the length of development and survival of *O. trimaculatus* embryos, and of three temperatures (13, 18 and 22 °C) and two salinities (30 and 33‰) on those of the zoeae I (Table I). Thermal treatments in set I included a variety of surface seawater conditions prevailing in the southern (colder) to the northern (warmer) limits of the range of geographical distribution of the species along the Atlantic coast of South America (Vinuesa 2005, Baldoni et al. 2015, Severino-Rodrigues et al. 2018), in combination with two salinity

Table I. Experimental design. Set I of experiments (shadowed cells): Testing length of development, survival and fitness of embryos (7 temperatures x 2 salinities) and zoeae I (3 temperatures x 2 salinities) of *Ovalipes trimaculatus*. Set II of experiments (thick framed cells): Testing length of development, survival and fitness of embryos and Zoeae I (1 temperature x 4 salinities) of *Ovalipes trimaculatus*.

	Temperature (ºC)							
		6	**10**	**13**	**15**	**18**	**22**	**24**
Salinity (‰)	26			e (7) z (3)				
	30	e (7)	e (7)	e (7) z (3)	e (7)	e (7) z (3)	e (7)	e (7) z (3)
	33	e (7)	e (7)	e (7) z (3)	e (7)	e (7) z (3)	e (7)	e (7) z (3)
	37			e (7) z (3)				

e: embryos; z: zoeae I; numbers in parenthesis indicate replicates in experiments. Each embryo replicate represents 50 embryos taken from one out of seven different ovigerous females. Each zoeae I replicate includes 100 zoeae I hatching from one out of three different ovigerous female egg masses.

treatments, including the typical sea surface salinity (33 ‰) on the inner shelf throughout most of its latitudinal distribution south of the mouth of La Plata river, and a lower salinity condition (30‰) common in shelf waters near the La Plata and other river mouths (Baldoni et al. 2015, Fenco Chavesta 2018). The second set tested the combination of four salinity and one thermal conditions, and was designed to establish the effects of saline stress on the same biological variables as in set I (Table I). In this, the lowest salinity (26‰) represented a condition present in estuaries at the mouth of the main rivers along the latitudinal distribution of *O. trimaculatus*, and the highest salinity condition (37‰) was slightly higher than that found in surface seawater at its northernmost limit (Ferreira et al. 1999).

Ovigerous females carrying embryos at the first stage of development (Martelli et al. 2016) were placed together in groups of 2 or 3 in 100-liter capacity lidded aquaria (85 × 85 × 60 cm; 2 aquaria per each treatment), provided with sand bottom, continuous aeration and refuges, and were feed *ad libitum* with fresh Argentinean hake (*Merluccius hubbsi*) meat. About 20% of seawater volume in the tanks was renewed daily, preserving the environmental variables from each treatment.

Daily, 50-eggs were taken with thin surgical pincers from the inner ovigerous mass of every female in each experimental treatment, placed in seawater on excavated slides and examined *in vivo* under an Olympus stereomicroscope CH30 (Tokyo, Japan) with total magnification of 100x. Based on changes in morphological and physiological characters described by Martelli et al. (2016), the stage of development and embryonic survival (S) were observed in the samples every day. Finally, the length of embryogenesis (L), counted in days from the date of collection of each ovigerous female carrying eggs at the first stage of development to the date of hatching was registered. Also, temperature necessary to complete the embryogenesis was estimated as:

$$DAT = \sum_{i=t_0}^{i=t_h} T\,^{\circ}C_i,$$

where DAT is the daily accumulated temperature, t_0 is the day of fertilization, t_h is the hatching day, and T°C is the average temperature (in °C) of each day during embryogenesis, assuming that fertilization occurred in the day

of capture of ovigerous females carrying the embryos (Barón 2002).

Artemia persimilis cysts (Biosima®) were disinfected with a sodium hypochlorite solution (5 mg L-1 of active chlorine) for 10 minutes, washed and incubated in sterile diluted seawater at 35°C and 15 g L-1 salinity under 2000-lux illumination and continuous aeration until hatching (Lavens & Sorgeloos 1996). Newly hatched nauplii were enriched during 24 h with a mix of microalgae composed of equal proportions of *Nannochloropsis oculata*, *Tetraselmis suecica*, *Dunaliella salina*, *Isochrysis galbana* and *Chaetoceros gracilis* at a 10^5 cells ml^{-1} (Chakraborty et al. 2007, Martelli 2018). Three groups of 100 *O. trimaculatus* zoeae I, newly hatched from eggs carried by females held at 33‰ and 13 ºC, were separated in individual beakers to be used as the 3 replicates in each of the six thermo-haline treatments (13, 18 and 22 °C × 30 and 33‰). Zoeae were fed on 2-4 enriched *Artemia* nauplii ml^{-1} in addition to a mix of microalgae in the culture seawater (10^6 cells ml^{-1}). Every morning the zoeae were transferred into a new beaker using a 5 ml pipette, counted and staged based on their total length, presence of appendages and shape of the eyes, following the descriptions of Schoeman & Cochroft (1996). For each treatment, length of the zoea I stage (L, in days), survival (S) from hatching to the first moult, and number of zoeae II (n_{ZII}) obtained from the total number of zoeae I at the beginning of the experiment, were registered.

Motility was determined as a measure of larval fitness based on the speed of vertical displacements (Vd, cm seg^{-1}) of zoeae I in response to the light stimulus. After feeding for five days, 10 zoeae I were randomly chosen from those in each of the six thermo-haline treatments (13, 18 and 22 °C × 30 and 33‰), and placed in a rectangular glass column (15 × 10 × 1 cm height × length × width) previously filled with 150 ml of sterilized seawater conditioned at the same thermo-haline condition as the larval culture. The column was placed within a dark box equipped with a white 2000 lumens led-light set on the top. Taking into account their observed positive phototaxism and high photokinesis, each *O. trimaculatus* zoea I was left at the bottom of the column and light was immediately turned on. Time (in seconds) taken by the zoeae I to displace from the bottom to the top of the column was registered with a digital chronometer, and speed of vertical displacement (Vd) was calculated thereafter. Whenever a zoea did not reach the top after 3 minutes, the experiment was stopped.

Statistical analysis

Two-way and one-way analyses of variance (ANOVA) were used to test mean differences between treatments for each variable of all experiments whenever normality (Kolmogorov-Smirnov test) and homoscedasticity (Fisher test) assumptions were fulfilled. A square root transformation was applied to analyze Vd data. In all cases, when differences were significant ($P < 0.05$), Tukey post hoc tests were used for treatment comparisons. Differences of slopes between linear-regression models fitted to ln-ln transformed relationships of length of embryonic development on temperature were tested on working spreadsheets following the procedure developed by Sokal & Rohlf (1995).

RESULTS

Within 6-24 °C and 30-33‰ (Set I of experiments), the chronology of embryonic development remained the same in all treatments, following the pattern described by Martelli et al. (2016). Mean length of *O. trimaculatus* embryogenesis varied with seawater temperature but not

with salinity (Table II, Figure 1). At the lowest experimental temperature (6 ºC) embryogenesis lasted up to 63 days, and as temperature increased to the highest tested value (24 ºC) its length decreased exponentially down to 19 days (Figure 2). The slope of the regression line of the log-log transformed length of *O. trimaculatus* embryogenesis on temperature was significantly lower (Slopes test, P<0.001) than that fitted for several other portunids pooled together, demonstrating that the influence of temperature on this variable is less marked than in other members of the family (Figure 2). Mean DAT necessary to complete the embryonic development did not show significant differences between thermal treatments within 10-22 ºC (30‰) / 13-22 ºC (33‰) (Figure 1), averaging 544.39 (± 60.43) ºC and 508.17 (± 47.61) ºC (mean ± SD) respectively for these ranges. Mean DAT necessary to complete the embryogenesis of the species was significantly lower in incubations at 6 ºC (30 and 33‰) (Figure 1).

No significant effects of salinity were observed on the mean survival of embryos incubated at 33 and 30‰ salinity (Table II). For those incubated at 33‰ salinity, mean survival did not differ between thermal treatments, except for 24 ºC where significantly fewer embryos survived. For embryos incubated at 30‰ salinity however, significantly higher mean survival was

Table II. Two-way ANOVA's for different variables of embryos and zoeae of *Ovalipes trimaculatus* subjected to different thermo-haline treatments within 6-24 ºC and 30-33‰.

	F.V.	SC	GL	CM	F	p
Length of embryogenesis	Temp	10663.54	6	1777.26	61.22	**<0.0001**
	Sal	5.99	1	5.99	0.21	0.6509
	Temp*Sal	115.86	6	19.31	0.67	0.6779
Length of Zoea I stage	Temp	57.26	1	57.26	87.26	**<0.0001**
	Sal	489.29	2	244.64	372.81	**<0.0001**
	Temp*Sal	21.54	2	10.77	16.41	**<0.0001**
Embryonic survival	Temp	2841.17	6	405.88	26.88	**0.0001**
	Sal	8.13	1	8.13	0.54	0.466
	Temp*Sal	499.74	6	99.95	6.62	**0.0001**
Survival of Zoea I stage	Temp	356.78	2	178.39	1.67	0.2286
	Sal	18.0	1	18.0	0.17	0.6884
	Temp*Sal	507.0	2	253.5	2.38	0.1349
nZII	Temp	3448.65	2	1724.3	19.56	**<0.0001**
	Sal	2792.93	1	2792.93	31.68	**<0.0001**
	Temp*Sal	115.02	2	57.54	0.65	**<0.0001**
Vd	Temp	0.97	2	0.48	8.33	**<0.0001**
	Sal	2.54	1	2.54	43.78	**0.005**
	Temp*Sal	0.20	2	0.14	2.49	**<0.0001**

Note: Bold letters indicate significant differences. nZII: number of zoeae achieving instar II relative to the number of zoeae I at the beginning of the experiment; Vd: speed of vertical displacements of zoeae I (cm seg-1) in response to light stimulus.

observed at mid-range temperatures (13, 15 and 18 ºC) (Figure 3).

The mean length of the *O. trimaculatus* zoea I stage was significantly shorter at 18 and 22 °C than at 13 °C (Table II, Figure 4). Both at 13 and 18 °C, mean length of zoea I stage was shorter at 30 than at 33‰ salinity (Figure 4). Both at 30 and 33‰, nZII was significantly higher at 18 and 22 °C compared to 13 °C (Figure 4). At 13 and 18 °C, mean nZII was significantly higher in incubations at 30 than at 33‰ salinity. At 18 and 22 °C, mean Dv was significantly higher in incubations at 30 than at 33‰. Survival of zoeae I did not show significant differences between tested treatments (Table II), averaging 54.75% ± 8.93% (mean ± SD).

For the combinations in Set II of experiments, salinity had significant effects on all measured variables except length of embryogenesis (Table III). Both embryos and zoeae I displayed significantly higher mean survival, nZII and Vd at 30 and 33‰ salinities compared to 26 and 37‰ (Figure 5). No zoea I molted to zoea II when incubated at 37‰ (Figure 6; Table III). The 30‰ salinity treatment resulted in significantly higher mean nZII than the others (Figure 6). Also, mean motility (Vd) was significantly higher and mean length of the zoea I stage was significantly shorter at this experimental condition (Figure 6).

DISCUSSION

Integration of the results of sets I and II of experiments in this work shows that the length of *O. trimaculatus* embryogenesis decreases exponentially as temperature increases from 6° (63 d) to 24 ºC (19 d), in agreement with the pattern observed in *Liocarcinus holsatus*, *Necora puber*, *Scylla serrata* and other portunids (Valdés et al. 1991, Hamasaki 2003).

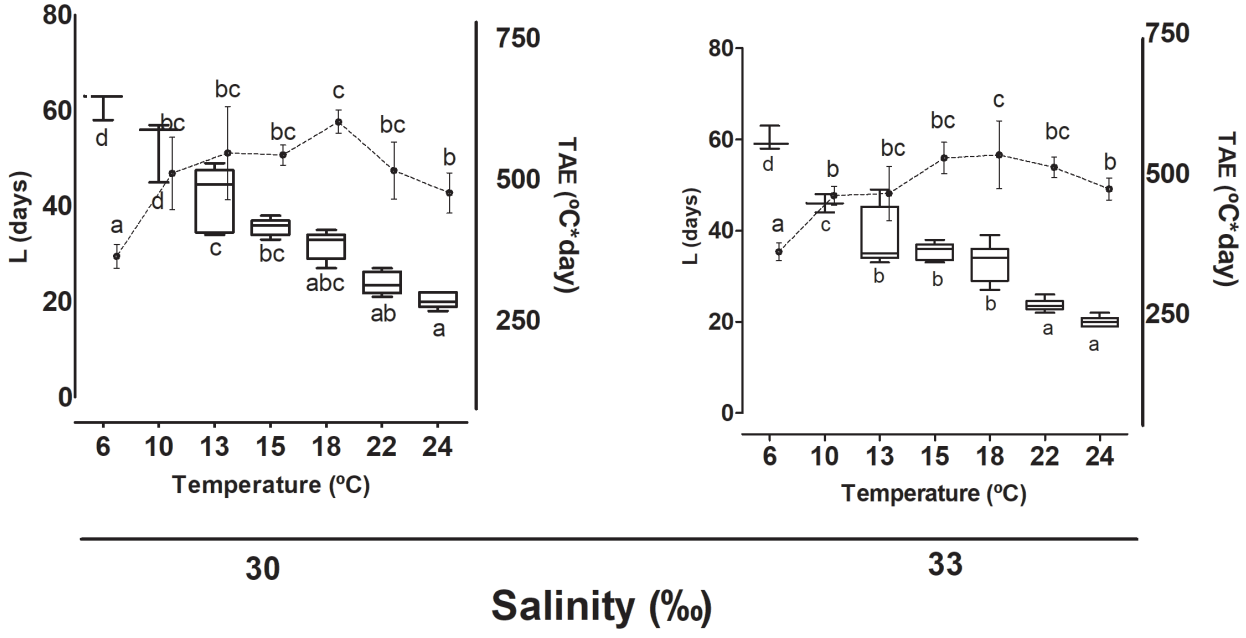

Figure 1. Length of *Ovalipes trimaculatus* embryogenesis (L, left Y axis) at different seawater temperature and salinity conditions and Daily Accumulated Temperature (DAT, right Y axis), representing the sum of daily average temperatures (°C) from fecundation to hatching. Different letters indicate significant statistical differences between means using Tuckey tests. Central mark in box plots represent the mean value; upper and lower box extremes show the inter-quartile range of observed values; whiskers show maximum and minimum observed values.

However, the embryogenesis of *O. trimaculatus* is markedly longer than that reported for most portunids with tropical geographical distribution, whose broodstocks are cultivated at higher temperatures, such as *Portunus sanguinolentus* (8-11 d at 28-31 ºC) (Samuel & Soundarapandían 2009), *P. pelagicus* (6-7 d at 28-31 ºC) (Soundarapandían & Tamizhazhagan 2009), *Scylla serrata* (7-9 d at 27-30 ºC) (Churchill 2003), *S. tranquebarica* (8.7 ± 0.6 d at 28.2 ± 0.2 ºC), *S. olivacea* (8.6 ± 0.2 d at 28.2 ± 0.1 ºC) (Ates et al. 2012) and *Arenaeus cribarius* (13.5 ± 2.1 d at 25.0 ± 2.0 ºC) (Pinheiro & Franzoso 2002). No significant effect of salinity on the length of embryogenesis of *O. trimaculatus* was detected either between 30 and 33‰ (all tested temperatures) or between 26, 30, 33 and 37‰ (13 ºC), in contrast to that reported for *Callinectes sapidus*, for whom it has been reported that the embryonic development is briefer at salinity conditions lower than normal values from oceanic waters (Costlow & Bookhout 1959).

Up to present, no information was available on the thermo-haline range enabling to complete the embryonic development of *O. trimaculatus*. Considering its wide latitudinal distribution (~47°-24° S) (Vinuesa 2005, Severino-Rodrigues et al. 2018) and its almost-exclusively-marine mode of life, experimental temperature and salinity values tested in this study were selected based on the hypothesis that the early life stages of the species are eurythermic and stenohaline as the juveniles and adults forms (Du Preez & McLachlan 1984, Martelli et al. 2019). In agreement, observed embryonic survival did not significantly differed between thermal treatments except for warmest condition (i.e., 24 ºC), at which significant temperature-salinity interaction was observed. Similarly, hatching of other portunids can be completed at relatively

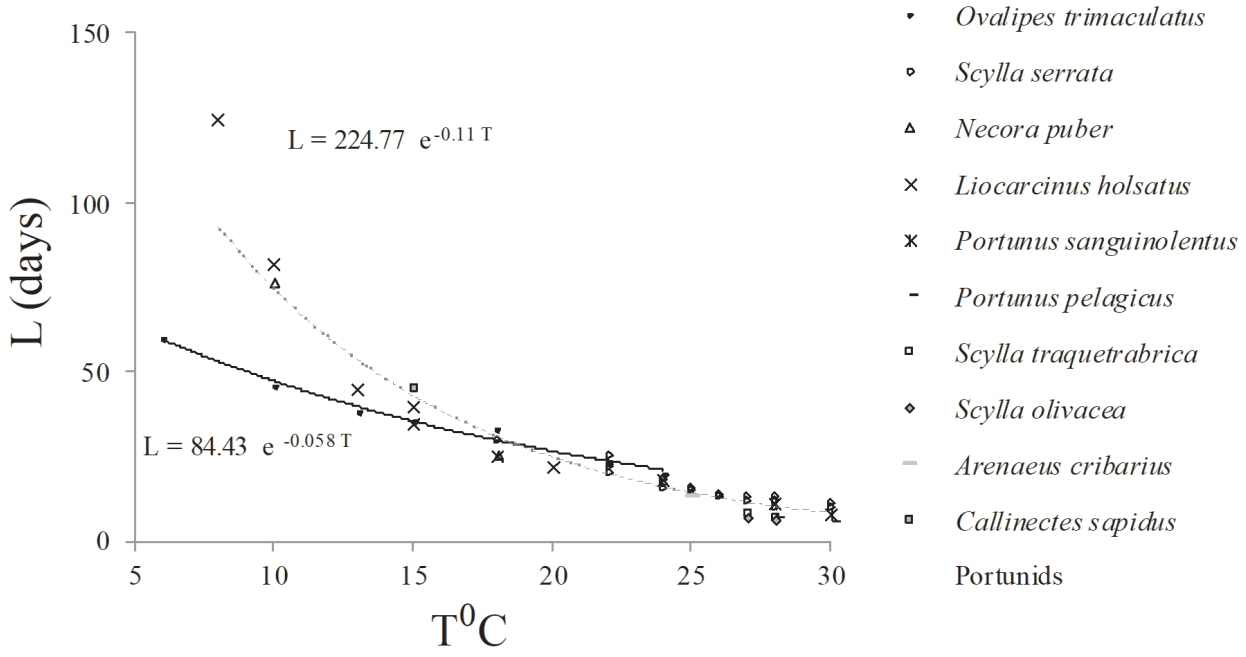

Figure 2. Length of embryogenesis of portunid crabs (L) at different seawater temperatures. The black solid line represents an exponential regression model fitted to data on the length of *Ovalipes trimacul*atus embryogenesis, and the grey dashed line represents an exponential regression model fitted to data on several other portunids pooled together (Choy 1991, Samuel & Soundarapandían 2009, Churchill 2003, Ates et al. 2012, Pinheiro & Franzoso 2002, Soundarapandían & Tamizhazhagan 2009, Amsler & George 1984).

wide temperature ranges, as for example in *C. sapidus* (19-29 ºC) (Jivoff et al. 2007), and in *Liocarcinus holsatus* and *Necora puber* (8–20 ºC) (Choy 1991). From all thermal conditions applied in this work, significantly different embryonic survival between the 30 and 33‰ salinity treatments were observed only at 24 ºC.

The osmoregulatory capacity in crustaceans relates to the presence and activity of the Na^+-K^+ and HCO_3-CL ATPases and carbonic anhydrase (Bas & Spivak 2000, Fukuda et al. 2017). While embryos of some species, generally those living permanently in oceanic waters, are osmoconformers, others living in environments with fluctuating salinity conditions develop osmoregulatory mechanisms (Charmantier & Charmantier-Daures 2001). Although many crustaceans acquire an osmoregulatory capability through the progress of ontogeny, others display it early in the embryogenesis (Miller et al. 2013, Vázquez et al. 2016). In some species of decapod crustaceans, osmoconformer embryos are protected from fluctuating external salinity by the outer egg membrane maintaining a hyper-osmotic condition at low salinity (Charmantier 1998, Stevens 2006). However, there is also evidence on the osmoregulatory activity of embryonic tissues/organs, like the ionocytes in the gills of *Astacus leptodactylus*, at least in the last phases of development (Charmantier & Charmantier-Daures 2001). In this work it was also observed that embryonic survival of *O. trimaculatus* is significantly lower at 26 and 37‰ compared to 30 and 33‰ (at 13 ºC), suggesting that the osmoregulatory capacity of the species is low during its embryogenesis, and that embryos are stenohaline osmoconformers. Likewise, hatching in some portunid species occurs in relatively narrow salinity ranges, as in *C. sapidus* (20 to 33‰, with an optimum within

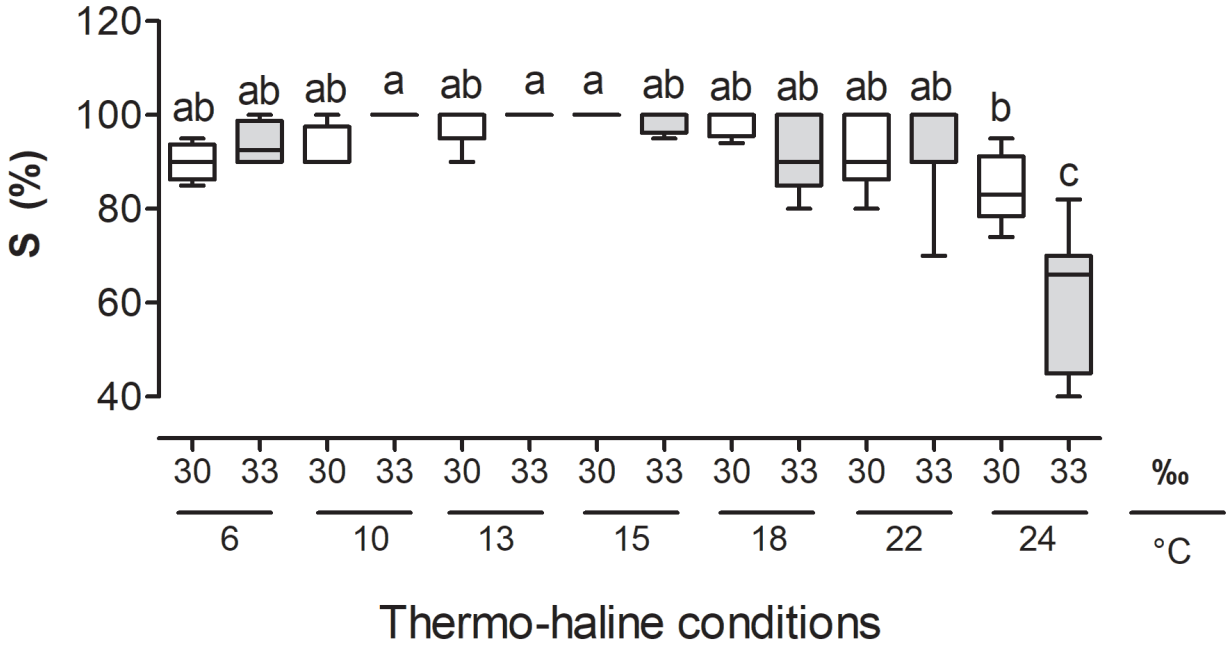

Figure 3. Survival of *Ovalipes trimaculatus* embryos incubated at different seawater temperature and salinity conditions. Different letters indicate significant statistical differences between mean survival (S) in Tuckey tests. Central mark in box plots represents the mean value; upper and lower box extremes show the inter-quartile range of observed values; whiskers show maximum and minimum observed values. Shadowed boxes indicate 33‰ salinity treatments.

23-28‰) (Costlow & Bookhout 1959) and *P. pelagicus* (25-45‰; survival >80% only within 30-35‰) (Noorulhudha et al. 2016). However, other portunids, as for example *Liocarcinus holsatus* and *Necora puber* (20-40‰), seem to have wider ranges of salinity tolerance during embryogenesis (Choy 1991). In addition, salinity conditions during embryogenesis can affect the survival and duration of larval stages, as observed in the burrowing crab *Neohelice granulata*, whose zoea I display higher initial carbon and nitrogen reserves, higher survival and shorter development time to zoea II when pre-hatching salinities are set at 15 and 20‰, as compared to 32‰ (Gimenez 2002).

From the perspective of aquaculture, it is worth to mention that incubations at 30‰ salinity combined with temperatures within 18-22 ºC may shorten the length of development of *O. trimaculatus* embryos and zoeae I respectively by 69% and 44% relative to their length at the prevailing mean thermo-haline condition during the reproductive season in the sampling area (33‰/13 ºC). However, considering that DAT (i.e., the sum of daily temperatures necessary to complete the embryonic development) did not show significant differences between 18-22 ºC and the other thermal treatments applied in this study (except for 6 ºC), it should be considered that broodstock or larvae incubation at that condition (i.e., 33‰/13 ºC) could result in zero energy expenditure in thermal regulation during hatchery operations solely at the expense of a longer incubation periods. On the other hand, since DAT showed no significant differences between most thermal treatments within 10-24 ºC, its overall mean value (438.91 ± 46.19 ºC , mean ± SD) can be effectively used in modelling larval recruitment patterns at different locations of the range of geographical distribution of *O. trimaculatus* accounting for seasonal and inter-annual variations of seawater temperature, as

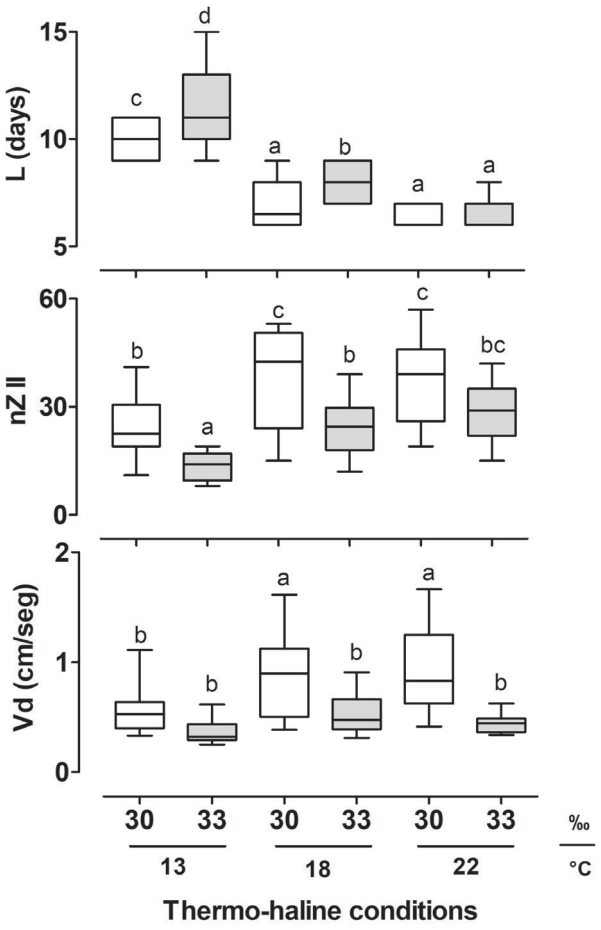

Figure 4. Length of *Ovalipes trimaculatus* Zoea I stage (L); number of zoeae achieving instar II relative to the number of zoeae I at the beginning of the experiment (nzII); speed of vertical displacements of the zoeae I (cm seg-1) in response to light stimulus (Vd) at different seawater temperature and salinity conditions. Different letters indicate significant statistical differences between mean L, nzII and Vd in Tuckey tests. Central mark in box plots represents the mean value; upper and lower box extremes show inter-quartile range of observed values; whiskers show maximum and minimum observed values. Shadowed boxes indicate 33‰ salinity treatments.

in previous studies conducted on other marine invertebrates (Barón 2002, Crespi-Abril & Barón 2012). Nevertheless, it should be considered that the length of embryogenesis and also DAT may vary between individuals from populations of the same species at the extremes of temperature clines, which may distribute their fatty acide

reserves in few/larger or many/smaller eggs as an evolutionary adaptation to local climate conditions (Rosa et al. 2007).

As observed in this study, length of the *O. trimaculatus* zoea I stage was affected by significant interactive effects of temperature and salinity. The stage lasted significantly longer at 33 than 30‰ at 13° and 18 ºC, but not at 22 ºC. At 13 ºC the zoea I stage lasted significantly less at 30‰ than at 26 and 33‰, and could not be completed at 37‰. Also, it lasted significantly longer at 13 ºC than 22 ºC (both at 30 and 33‰ salinities). In contrast, although *Necora puber* zoeae I stage lasts 7 d at 15 ºC, and 4 d at 20 ºC, no interaction was observed with salinity (25, 30 and 35‰) (Mene et al. 1991), while both at 25 and 35‰, the *Scylla tranquebarica* zoeae I stage lasts 7-8 d at 20 ºC, 3 d at 26 ºC and 2 d at 32 ºC (Baylon 2013).

Mean survival of *O. trimaculatus* zoeae I did not show significant differences between thermo-haline treatments (13, 18 and 22 ºC combined with 30 and 33‰). However, at 13 ºC it was significantly lower at 26 and 37‰ than at 30 and 33‰. In agreement, zoeae I of *O. trimaculatus* from South African coastal waters exposed for brief (24 h) periods to several thermo-haline conditions showed optimal survivals at 15-22 ºC, still relatively high (≥80%) at 7 ºC (30-35‰), but did not survived at temperatures above 27.5 ºC (Du Preez & McLachlan 1984). Similarly, zoeae I from *Charybdis japonica*, exposed to different thermo-haline treatments during 24 h, displayed high survival in thermal treatments within 16-25 ºC, still relatively high over a wider range (≥80% at 12–33 ºC), suggesting that its eurythermal character could have allowed this species to expand its distribution beyond its native range during the last two decades (Fowler et al. 2011). Also, *O. trimaculatus* zoeae I from the coast of South Africa showed negligible or null survival at salinities below 20‰, were active within 20-35‰ and 10.5-25 ºC, but only showed high survival (≥90%) at 10-23 ºC/30-35‰ (Du Preez & McLachlan 1984). In agreement, zoeae I of the portunid *Charybdis feriata* can complete their development within 20-40‰, but with high survival (≥ 90%) only at 35‰ (70% at 30‰ / 55% at 40‰) (Soundarapandian et al. 2013).

Within the range of thermal treatments applied in this study, *O. trimaculatus* zoeae I showed a marked reduction in the length of the stage and an increase in motility (Vd) as temperature and salinity approached to an optimum (i.e., 18-22° C/30‰), as reported for crustacean larvae in general (Putro & Fahrian 2015). Also, nZII was affected by the significant interaction of temperature and salinity (at least for 13, 18 and 22 ºC combined with 30 and 33‰), lowest and highest nZII occurring at 13 ºC/33‰ and 18-22 ºC/30‰ respectively.

Since in the sampling area of this study, early zoeae of *O. trimaculatus* are found from Oct-Nov to Feb-Mar, when surface seawater (~33‰) has temperatures varying within 13-18° C (Dellatorre et al. 2014), experimental results from this study should allow accelerating their rearing by a slight increase of seawater temperature up to 18-22° C and a subtle reduction of salinity down to 30‰, maintaining high larval fitness and survival and reducing aquaculture production costs (Moksnes et al. 2015, Quinitio 2015). Also, these will provide a deeper understanding of the life cycle of the species in the sampling area and along its latitudinal distribution in the Atlantic coast of South America, useful for the design of management rules and to develop numerical models allowing to predict the timing and success of larval recruitment at a spatio-temporal scale.

Table III. One-way ANOVA's for different variables registered for the embryonic and first larval stage of *Ovalipes trimaculatus* as response to different seawater salinities.

F.V.	SC	GL	CM	F	P
Embryonic survival	4140.12	3	1380.04	21.76	**<0.0001**
Survival of Zoea I stage	1791.38	2	895.69	69.43	**0.001**
Length of embryogenesis	1,16	3	0,39	0,61	0,3182
Length of zoea I stage	14.77	2	7.38	7.09	**0.0013**
nzII	9856.60	3	3285.53	37.15	**<0.0001**
Vd	7.19	3	2.4	47.54	**<0.0001**

Note: Bold letters indicate significant differences. nZII: number of zoeae achieving instar II relative to the number of zoeae I at the beginning of the experiment; Vd: speed of vertical displacements of zoeae I (cm seg-1) in response to light stimulus.

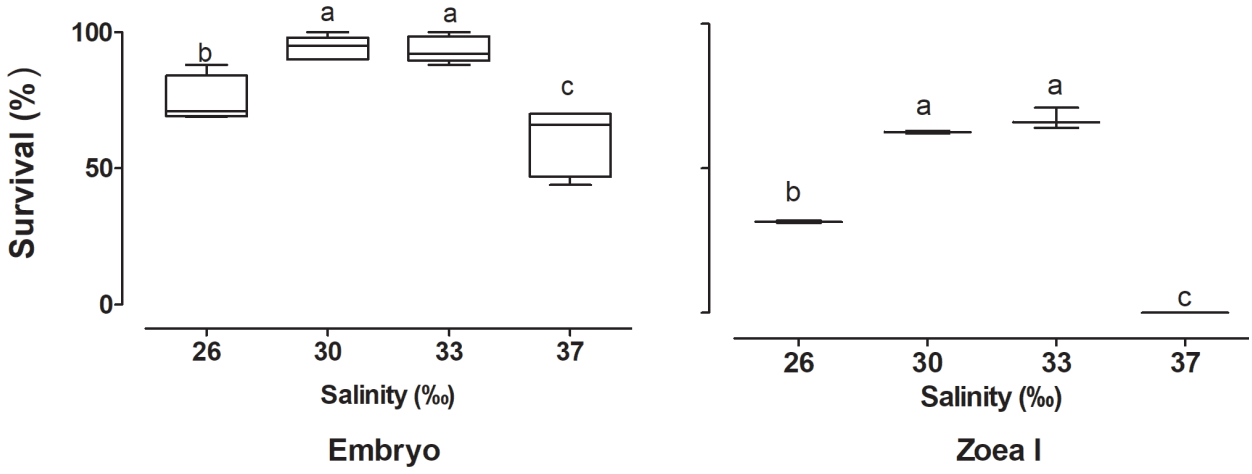

Figure 5. *Ovalipes trimaculatus* survival in embryos and zoeae I at different seawater salinity conditions. Different letters indicate significant statistical differences between mean survival (S) in Tuckey test. Central mark in box plots represents the mean value; upper and lower box extremes show the inter-quartile range of observed values; whiskers show maximum and minimum observed values.

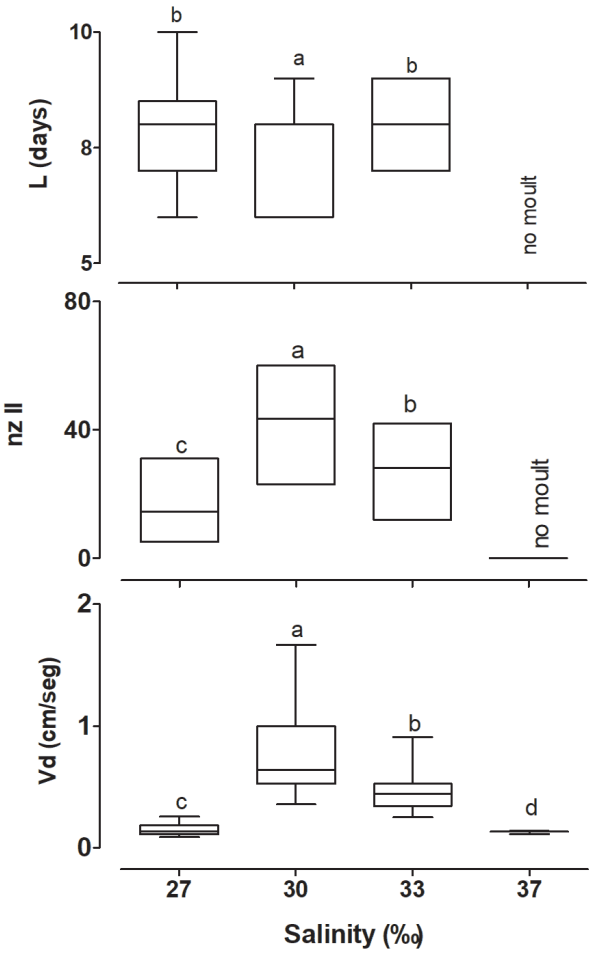

Figure 6. Length of *Ovalipes trimaculatus* Zoea I stage (L); speed of vertical displacements of zoeae I (cm seg-1) in response to light stimulus (Vd); number of zoeae achieving instar II relative to the number of zoeae I at the beginning of the experiments (nzII) at different salinity conditions. Different letters indicate significant statistical differences between mean L, nzII and Dv in Tuckey tests. Central mark in box plots represents the mean value; upper and lower box extremes show the inter-quartile range of observed values; whiskers show maximum and minimum observed values.

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REFERENCES

ALLAN G & FIELDER. 2004. Mud crab aquaculture in Australia and Southeast Asia. Proceedings of a Scoping Study and Workshop. ACIAR 54.

AMSLER MO & GEORGE RY. 1984. The effect of temperature on the oxygen consumption and developmental rate of the embryos of *Callinectes sapidus* Rathbun. J Exp Mar Biol Ecol 82(2-3): 221-229.

ANGER K. 2001. The biology of decapod crustacean larvae. In Lisse: AA Balkema Publishers 14: 1-420.

ATES MCD, QUINITIO ET & SANARES RC. 2012. Comparative study on the embryonic development of three mud crabs *Scylla spp*. Aquac Res 43(2): 215-225.

BALDONI A, MOLINARI GN, RETA R & GUERRERO RA. 2015. Atlas de temperatura and salinidad de la plataforma continental del Atlántico Sudoccidental: períodos cálido y frío. Mar del Plata: Instituto Nacional de Investigación and Desarrollo Pesquero INIDEP 85.

BARON PJ. 2002. Embryonic development of *Loligo gahi* and modeling of hatching frequency distributions in Patagonia. Bull Mar Sci 71(1): 165-173.

BAS CC & SPIVAK ED. 2000. Effect of salinity on embryos of two southwestern Atlantic estuarine grapsid crab species cultured *in vitro*. J Crustac 20(4): 647-656.

BAS CC, SPIVAK ED & ANGER K. 2008. Variation in early developmental stages in two populations of an intertidal crab, *Neohelice* (*Chasmagnathus*) *granulata*. Helgol Mar Res 62(4): 393.

BAYLON JC. 2013. The combined effects of salinity and temperature on the survival and development of zoea, megalopa and crab instar larvae of mud crab, *Scylla tranquebarica* (Fabricius 1798). Asian Fish Sci 26(1): 14-25.

BIRCHARD GF, DROLET L & MANTEL LH. 1982. The effect of reduced salinity on osmoregulation and oxygen consumption in the lady crab, *Ovalipes ocellatus* (Herbst). Comp Biochem Physiol B: Physiol 71: 321-324.

BOYKO CB & LIGUORI A. 2014. Swimming Crabs (Crustacea: Brachyura: Portunoidea) of Easter Island. Pac Sci 68(4): 563-576.

BURGUENER MG & BARÓN PJ. 2017. Prospectiva Tecnológica en Maricultura: Argentina en Contexto Global. I. Vigilancia Tecnológica. Editorial Académica Española. Saarbrücken, Germany, 173 p.

CASTEJÓN D, ROTLLANT G, GIMÉNEZ L, TORRES G & GUERAO G. 2015. The effects of temperature and salinity on the survival, growth and duration of the larval development of the common spider crab *Maja brachydactyla* (Balss, 1922) (Brachyura: Majidae). J Shell Res 34(3): 1073-1083.

CHAKRABORTY RD, CHAKRABORTY K & RADHAKRISHNAN EV. 2007. Variation in fatty acid composition of *Artemia salina* nauplius enriched with microalgae and baker's yeast for use in larviculture. J Agric Food Chem 55(10): 4043-4051.

CHARMANTIER G. 1998. Ontogeny of osmoregulation in crustacean: a review. Inverteb Reprod Dev 33: 177-190.

CHARMANTIER G & CHARMANTIER-DAURES M. 2001. Ontogeny of osmoregulation in crustaceans: the embryonic phase. Am Zool 41(5): 1078-1089.

CHURCHILL GR. 2003. An Investigation into the captive spawning, egg characteristic and egg quality of the mud crab *(Scylla serrata*) in South Africa. Workshop of mud crab culture, ecology and fisheries, Can Tho University, Vietnam, 8-10th January.

CINTI A. 2002. El efecto de factores ambientales en la supervivencia embrionaria del calamar patagónico *Loligo gahi*. Tesis de Licenciatura. Universidad Nacional de la Patagonia San Juan Bosco (UNPSJB), Puerto Madryn, Chubut, Argentina.

COSTLOW JR & JDOOKHOUT CG. 1959. The larval development of Callinectes sapidus Rathbun reared in the laboratory. Biol Bull 116(3): 373-396.

CRESPI-ABRIL AC & BARÓN PJ. 2012. Revision of the population structuring of *Illex argentinus* (Castellanos, 1960) and a new interpretation based on modelling the spatio-temporal environmental suitability for spawning and nursery. Fish Oceanogr 21(2-3): 199-214.

DE LA BARRA P. 2018. Ecología trófica y análisis de la pesquería del cangrejo nadador *Ovalipes trimaculatus* en el norte del Golfo San Matías. Facultad de Ciencias Exactas y Naturales. Universidad de Buenos Aires.

DELLATORRE FG, LOVRICH G, ROJAS L, MILANO V, FIGUEROA ND & BARÓN PJ. 2014. Seasonal abundance and vertical distribution of crab larvae from northern Patagonia (Argentina): implications for their transport in a biogeographic boundary region. Mar Biol Res 10(1): 37-50.

DELLATORRE FG, PISONI JP, BARÓN PJ & RIVAS AL. 2012. Tide and wind forced nearshore dynamics in Nuevo Gulf (Northern Patagonia, Argentina): potential implications for cross-shore transport. J Mar Sys 96: 82-89.

DIMA JB, BARÓN PJ & ZARITZKY NE. 2012. Mathematical modeling of the heat transfer process and protein denaturation during the thermal treatment of Patagonian Marine Crabs. J Food Eng 113(4): 623-634.

DIMA JB, BARÓN PJ & ZARITZKY NE. 2016. Pasteurization conditions and evaluation of quality parameters of frozen packaged crab meat. J Aquat Food Prod Technol 25(5): 745-759.

DIMA JB, SANTOS MV, BARÓN PJ, CALIFANO A & ZARITZKY NE. 2014. Experimental study and numerical modeling of the freezing process of marine products. Food Bioprod Process 92: 54-66.

DISALVO LH, RANDALL JE & CEA A. 1988. Ecological reconnaissance of the Easter Island sublittoral marine environment. Nat Geogr Res 4(4): 451-473.

DOHLE W, GERBERDING M, HEJNOL A & SCHOLTZ G. 2004. Cell lineage, segment differentiation, and gene expression in crustaceans. In: Scholtz G.(Ed) Evol Develop Biol Crust 5: 95-133.

DU PREEZ HH & MCLACHLAN A. 1984. Biology of the three spot swimming crab, *Ovalipes punctatus* (De Haan) III. Reproduction, fecundity and egg development. Crustaceana 285-297.

FENCO CHAVESTA HA. 2018. Observaciones satelitales de salinidad superficial en el Océano Atlántico Sudoccidental: Intercambios entre plataforma y talud. Tesis de Maestría en Geomática, Universidad Nacional de La Plata, 108 p.

FERREIRA MM, FARIA CG & PAES ET. 1999. Oceanographic characterization of northern Sao Paulo Coast: a chemometric study. Chemom Intell Lab Syst 47(2): 289-297.

FOWLER AE, GERNER NV & SEWELL MA. 2011. Temperature and salinity tolerances of Stage 1 zoeae predict possible range expansion of an introduced portunid crab, Charybdis japonica, in New Zealand. Bioll Invasions 13(3): 691-699.

FUKUDA B, BERTINI G & ALMEIDA LCFD. 2017. Effect of salinity on the embryonic development of *Macrobrachium acanthurus* (Decapoda: Palaemonidae). Invertebr Reprod Dev 61(1): 1-8.

GARCÉS-VARGAS J, RURIZ M, PARDO LM, NUÑEZ S & PÉREZ-SANTOS I. 2013. Caracterización hidrográfica del estuario del río Valdivia, centro-sur de Chile. Lat Am J Aquat Res 41(1): 113-125.

GIMENEZ L. 2002. Effects of prehatching salinity and initial larval biomass on survival and duration of development in the zoea 1 of the estuarine crab, *Chasmagnathus granulata*, under nutritional stress. J Exp Mar Biol Ecol 270(1): 93-110.

GIMÉNEZ L & ANGER K. 2001. Relationships among salinity, egg size, embryonic development, and larval biomass in the estuarine crab *Chasmagnathus granulata* Dana, 1851. J Exp Mar Biol Ecol 260(2): 241-257.

GONG J, YU K, SHU L, YE H, LI S & ZENG C. 2015. Evaluating the effects of temperature, salinity, starvation and autotomy on molting success, molting interval and expression of ecdysone receptor in early juvenile mud crabs, *Scylla paramamosain*. J Exp Mar Biol Ecol 464: 11-17.

HAMASAKI K. 2003. Effects of temperature on the egg incubation period, survival and developmental period of larvae of the mud crab *Scylla serrate* (Forskål) (Brachyura: Portunidae) reared in the laboratory. Aquac 219(1): 561-572.

IKHWANUDDIN M, HAYIMAD T, GHAZALI A, HALIM SSA & ABDULLAH SA. 2016. Resistance test at early larval stage of blue swimming crab, *Portunus pelagicus*. Songklanakarin J Sci Tech 38(1).

JIVOFF PR, HINES AH & QUACKENBUSH S. 2007. Reproduction and embryonic development. In Blue crab: *Callinectes sapidus*.

LAVENS P & SORGELOOS P. 1996. Manual on the production and use of live food for aquaculture (No. 361). Food and Agriculture Organization (FAO).

LEZCANO AH, ROJAS QUIROGA ML, LIBEROFF AL & VAN DER MOLEN S. 2015. Marine pollution effects on the southern surf crab *Ovalipes trimaculatus* (Crustacea: Brachyura: Polybiidae) in Patagonia Argentina. Mar Pollut Bull 91(2): 524-529.

LUPPI TA, SPIVAK ED & BAS CC. 2003. The effects of temperature and salinity on larval development of *Armases rubripes* Rathbun, 1897 (Brachyura, Grapsoidea, Sesarmidae), and the southern limit of its geographical distribution. Estuar Coast Shelf Sci 58(3): 575-585.

MARTELLI A. 2018. Evaluación del desarrollo de las etapas tempranas del ciclo de vida del cangrejo nadador, *Ovalipes trimaculatus* (de Hann 1833) y su aplicación en acuicultura. Universidad Nacional del Comahue.

MARTELLI A, HERNANDEZ MR, BARKI L & BARON PJ. 2019. The effect of temperature, salinity, eyestalk ablation, and limb removal on growth and survival of juvenile paddle crab, *Ovalipes trimaculatus* (Brachyura: Portunidae). Bull Mar Sci 95(1): 69-82.

MARTELLI A, TAPELLA F, GONZÁLEZ-PISANI X, DELLATORRE F & BARON PJ. 2016. Embryonic development of the southern surf crab *Ovalipes trimaculatus* (Decapoda: Brachyura: Portunoidea). Sci Mar 80(4): 499-509.

MELO GAS. 2010. The brachyura (crustacea: decapoda) collected by the gedip project between Torres, Rio Grande do Sul (Brazil) and Maldonado (Uruguay).

MENE L, ALVAREZ-OSSORIO MT, GONZALEZ-GURRIARAN E & VALDÉS L. 1991. Effects of temperature and salinity on larval development of *Necora puber* (Brachyura: Portunidae). Mar Biol 108(1): 73-81.

MILLER NA, PAGANINI AW & STILLMAN JH. 2013. Differential thermal tolerance and energetic trajectories during ontogeny in porcelain crabs, genus *Petrolisthes*. J Therm Biol 38(2): 79-85.

MOKSNES PO, MIRERA D, LOKINA R, OCHIEWO J, MAHUDI H, JIDDAWI N & TROELL M. 2015. Feasibility of extensive, small-scale mud crab farming (*Scylla serrata*) in East Africa. West Indían Ocean J Mar Sci 14(1-2): 23-38.

NOORULHUDHA NF, IKHWANUDDIN M, AZRA MN, SITI-AISHAH A & ABOL-MUNAFI AB. 2016. Embryonic development and hatching rate of blue swimming crab, *Portunus pelagicus* (Linnaeus, 1758) under different water salinities. Turkish J Fish Aquat Sci 16: 669-677.

NURDIANI R & ZENG C. 2007. Effects of temperature and salinity on the survival and development of mud crab, Scylla serrata (Forsskål), larvae. Aquac Res 38(14): 1529-1538.

PAPADOPOULOS I, NEWMAN BK, SCHOEMAN DS & WOOLDRIDGE TH. 2006. Influence of salinity and temperature on the larval development of the crown crab, *Hymenosoma orbiculare* (Crustacea: Brachyura: Hymenosomatidae). Afr J Aquat Sci 31(1): 43-52.

PARDO LM, RUBILAR PS & FUENTES JP. 2020. North Patagonian estuaries appear to function as nursery habitats for marble crab (*Metacarcinus edwardsii*). Reg Stud in Mar Sci 101315.

PINHEIRO M & FRANZOSO A. 2002. Reproduction of the speckled swimming crab *Arenaeus cribrarius* (Brachyura: Portunidae) on the Brazilian coast near 23°30'S. J Crust Biol 22(2): 416-428.

PUTRO SP & FAHRIAN HH. 2015. Application of Environmental Management on the Farming Practice of Mud Crab *Scylla serrata* at Coastal Area of Ujung Alang, Cilacap, Indonesia: Efforts toward Sustainable Aquaculture. Procedía Environ Sci 23: 297-306.

QUINITIO ET. 2015. Status of mud crab industry in the Philippines. In Proceedings of the International Seminar-Workshop on Mud Crab Aquaculture and Fisheries Management, 10-12 April 2013, Tamil Nadu, India. Rajiv Gandhi Centre for Aquaculture (MPEDA), 27-35 p.

RAHMAN MM, ISLAM MA, HAQUE SM & WAHAB A. 2017. Mud Crab Aquaculture and Fisheries in Coastal Bangladesh. World Aquac 47.

RIVAS A & RIPA P. 1989. Variación estacional de la estructura termo-halina de golfo Nuevo, Argentina. Rev Geof 28: 3-23.

ROSA R, CALADO R, NARCISO L & NUNES ML. 2007. Embryogenesis of decapod crustaceans with different life history traits, feeding ecologies and habitats: a fatty acid approach. Mar Biol 151(3): 935-947.

SAINZ-HERNÁNDEZ JC, FIERRO-CORONADO JA, AGUIÑAGA-CRUZ JA, GARCÍA-RODRIGUEZ LD, BARRAZA-LÓPEZ JS, SANTAMARÍA-MIRANDA A & CASTRO-MARTÍNEZ C. 2016. Effect of temperature on the morphometric development of eggs in the prawn *Macrobrachium americanum* (Caridea: Palaemonidae) and larval success under experimental conditions. Inverteb Reprod Dev 60(3): 194-200.

SAMUEL NJ & SOUNDARAPANDÍAN P. 2009. Embryonic development of commercially important portunid crab *Portunus sanguinolentus* (Herbst). Int J Anim Vet Adv 1(2): 32-38.

SANTOS MV, DIMA JB, BARÓN PJ, CALIFANO AN & ZARITZKY NE. 2012. Simulación numérica del proceso industrial de congelación de cangrejos patagónicos. Mec Comput 31: 2133-2149.

SCHIFFER M, HARMS L, LUCASSEN M, MARK FC, PÓRTNER HO & STORCH D. 2014. Temperature tolerance of different larval stages of the spider crab *Hyas araneus* exposed to elevated seawater P CO_2. Front Zool 11(1): 87.

SCHOEMAN DS & COCKCROFT AC. 1993. On the misidentification of a common sandy beach crab belonging to the genus *Ovalipes* Rathbun, 1898. Afr Zool 28(2): 124-125.

SCHOEMAN DS & COCKROFT AC. 1996. A Description of *Ovalipes trimaculatus* (De Hann 1833) (Brachyura: Portunidae: Polybiinae) zoeae reared in the laboratory. S Afr J Mar Sci 17: 113-134.

SCHOLTZ G & WOLFF C. 2013. Arthropod embryology: cleavage and germ band development. In Arthrop Biol Evol 63-89.

SEVERINO-RODRIGUES E, DAS CHAGAS SOARES F, DA GRACA-LOPES R, DE SOUZA KH & CANEO VOC. 2018. Diversidade e biologia de espécies de portunidae (Decapoda, Brachyura) no estuário de Iguape, Ilha Comprida e Cananéia, São Paulo, Brasil. Boletim do Instituto de Pesca 35(1): 47-60.

SOKAL RR & ROHLF FJ. 1995. Biometry: the principles and practice of statistics in biological research, 68-70.

SOUNDARAPANDIAN P, ILAVARASAN N, VARADHARAJAN D, KUMAN J & SURESH B. 2013. Embryonic development of commercially important Portunid crab *Charybdis feriata* (Linnaeus). J Mar Sci: Res Develop 3: 122.

SOUNDARAPANDIAN P & TAMIZHAZHAGAN T. 2009. Embryonic development of commercially important swimming crab *Portunus pelagicus* (Linnaeus). Curr Res J Biol Sci 1(3): 106-108.

STEVENS BG. 2006. Embryo development and morphometry in the blue king crab *Paralithodes platypus* studied by using image and cluster analysis. J Shell Res 25(2): 569-576.

TANKERSLEY RA & FORWARD JR RB. 2007. Environmental physiology. The Blue Crab: *Callinectes sapidus*. Maryland Sea Grant, College Park, MD, 451-484.

VALDES L, ALVAREZ-OSSORIO MT & GONZALEZ-GURRIARÁN E. 1991. Incubation of eggs of *Necora puber* (L.,1767) (Decapoda, Brachyura, Portunidae). Volume and biomass changes in embryonic development. Crustaceana 60(2): 163-177.

VÁZQUEZ MG, BAS CC & SPIVAK ED. 2016. Ontogeny of salinity tolerance in the invasive shrimp *Palaemon macrodactylus (Caridea: Palaemonidae*). J Crust Biol 36(2): 214-219.

VINUESA J. 2005. Distribución de crustáceos decápodos and estomatópodos del Golfo San Jorge, Argentina. Rev Biol Mar Ocean 40: 7-21.

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A. Martelli: Collected all data, analysis tools, wrote the paper and P. Baron: Conceived and designed the analysis, financial and advisor of all work.