Short Communication

Reproductive capacity of the red cusk-eel Genypterus chilensis (Guichenot, 1848) in captivity

Rolando Vega^{1,3}, Clara Sepúlveda¹, Maicol Barnert¹, Alfonso Mardones^{1,3} Francisco Encina-Montoya^{2,4}, Carlos Oberti², Diego Ramirez⁵ & Juan Manuel Estrada⁵ ¹Escuela de Acuicultura, Facultad de Recursos Naturales Universidad Católica de Temuco, Temuco, Chile ²Escuela de Ciencias Ambientales, Facultad de Recursos Naturales Universidad Católica de Temuco, Temuco, Chile ³Núcleo de Investigación de Producción Alimentaria Universidad Católica de Temuco, Temuco, Chile ⁴Núcleo de Investigación de Estudios Ambientales Universidad Católica de Temuco, Temuco, Chile ⁵CIMARQ, Universidad Andrés Bello, Quintay, Chile Corresponding author: Rolando Vega (rvega@uct.cl)

ABSTRACT. *Genypterus chilensis* is a marine fish of high gastronomic demand, whose capture has declined in recent years due to overfishing. In the development of the farming technology, high mortalities were obtained during egg incubation. The objective of this study is to contribute to the knowledge of fecundity and eggs viability of *G. chilensis* in captivity. The spawns of *G. chilensis* were analyzed over a period of 2 years and 3 months. The total fecundity was estimated by counting the masses and eggs produced monthly throughout the period. The results confirm that *G. chilensis* is a partial spawner, since a female may more than two masses of eggs per day, due to a large amount of mass spawned per season (621 average). The total production of masses of the Farming Centre during the period was 2,290; of these, only 7% (166) corresponding to 15,330,517 eggs were incubated. Because of its high fecundity, *G. chilensis* produces numerous masses of eggs, of which only a small percentage reaches incubation, as well as it occurs in other marine fish.

Keywords: Genypterus chilensis, reproductive capacity, fecundity, egg viability, aquaculture.

The red cusk-eel *Genypterus chilensis* (Guichenot, 1848) is a marine fish from the Ophidiidae family, and its common name comes from its reddish colored belly, throat, and lips. Its geographical distribution extends from Paita in Peru to Cape Horn in southern Chile (Kong *et al.*, 1988). It mainly inhabits in coastal bodies of water at depths that generally range between 20 to 150 m; it is a benthic species of the continental shelf usually found on rocky substrates, feeding on fish, crustaceans, and mollusks (Chong *et al.*, 2006).

G. chilensis has been a highly attractive resource for the local market due to the excellent quality of the meat, which has led to the overharvesting of this species over the last 12 years, producing a sharp decline in landing volumes. This situation has led to a critical level of supply shortage and a growing increase of unmet local demand. In 1994, the fishery of *Genypterus chilensis* generated a supply for about seven millions of consumers, however, nowadays the supply of this resource has been reduced, is barely enough for 400 thousand people (UNAB, 2012). In quantitative terms, the national landings decreased from 1,712 annual ton in 1994 to 636 ton in 2014, with a minimum of 213 ton in 2002 (Chong *et al.*, 2006; Chong & González, 2009; SERNAPESCA, 2016). As a consequence of the supply shortages, a gradual and accelerated increase in the prices of the product has been observed, registering a growing demand, estimated in about 3,048 ton a year, which has not been met until today (Vega *et al.*, 2012).

Many marine fishes (Merluccius hubbsi, M. gayi, Strangomera bentincki, Engraulis anchoita, Sardinops sagax, Paralichthys patagonicus, Genypterus chilensis,

Corresponding editor: Enrique Dupré

G. blacodes, G. maculatus and G. capensis) from cold temperate waters have broad annual spawning periods, with mass spawning in late winter and during spring, which are the most productive periods, when temperatures fluctuate between 12 and 14°C with a decrease or pause during summer (Olivar & Sabatés, 1989; Machinandiarena et al., 1998; Tascheri et al., 2003; Paredes & Bravo, 2005; Chong & González, 2009). These fishes are both multiple and partial spawners. between short intervals, producing thousands of eggs with high variability, being the species Dicentrarchus labrax an example of high fecundity with 300,000 eggs kg⁻¹ (Büke, 2002). G. blacodes fecundity increases with body weight and length, with an average of $333,333 \pm 241,642$ eggs per female between 0.3 to 12 kg of weight and 85 to 120 cm in total length, and with a highly variable range from 66,167 to 1,230,000 eggs per female (Paredes & Bravo, 2005).

In captivity, overnight *G. chilensis* spawns gelatinous masses of floating eggs of high fecundity which reach on average up to 80,000 eggs of 1.3 mm in diameter. The greater the size and age the greater the fertility, reaching a maximum of 285,000 eggs per mass. Depending on the temperature (15 to 18° C) incubation lasts 4 to 5 days, hatching pelagic larvae of 5 mm. During the first days of incubation the mass dissolves, eggs are released, floating the living ones and decanting the dead ones to the bottom (Vega *et al.*, 2012).

In fishes of high fecundity, greater than hundred thousand eggs, the viability of eggs is highly variable. A Paralichthys adspersus female in captivity can produce a mean of 2,125,000 eggs kg⁻¹ yearly, from which 30 to 50% are floating viable, with fecundity rates that range between 0 and 100%, per spawning. The highest percentage of viable eggs is obtained during a limited period of two months during spring and with a temperature of 14-15°C, although spawns may be obtained with a minimum temperature of 12.7°C and a maximum of 19.7°C. Non-viable eggs have an opaque appearance, with irregular diameter and surface, abnormal yolk distribution and occasionally with 2 or 3 lipid droplets, being present during the whole annual spawning period, but their number tends to increase when outside of the peak egg production period, when the temperature exceeds 16°C (Silva & Oliva, 2010).

In order to solve the problem of unmet demand of *G. chilensis*, one alternative is its production through fish farming. In the development of the farming technology, mass production of eggs has been achieved with high mortalities, mainly due the lack of knowledge of the reproductive capacity of the species, which leads to the goal of this study whose aim is to contribute to

deepen the knowledge of *G. chilensis* fecundity and egg viability through fish spawning in captivity.

The eggs from the brood stock spawned masses of the red cusk-eel, G. chilensis (Guichenot, 1848), from the Quintay Centre for Marine Research (CIMARQ), Andres Bello National University, Valparaiso, Chile, within the Ouintav area (33°11'S, 71°10'W), were analysed over a period of 27 months, from September 2007 to December 2009. 61 broodstock of wild origin were captured, weighed and kept in black coloured fiberglass tanks of 7 and 10 m³, with a water column height of 0.8 m, from where the volumes of the floating masses were daily extracted and measured and eggs were counted daily, according to the methodology described in Vega et al. (2012), which indicates capture, feeding methods of fishes in captivity and seawater physical and chemical parameters. Using a YSI oxygen meter, model 55-25 FT (Ohio, USA), measures of tank water oxygen concentration (mg L^{-1}), oxygen saturation percentage and temperature (°C) were daily taken at 10:00 and 16:00 h. The salinity of seawater was measured using ATC HI-9835 water parameter measurement equipment (RI, USA).

In January 2009, through an ultrasound scanner (BCF, Easy Scan model, NJ, USA), the gender of 24 specimens was identified, of which 11 were female and 13 were male; assuming that the sex ratio is 1:1, males were homogeneously distributed in five tanks in order to guarantee the fertilization of eggs. It is assumed that the 37 specimens of unidentified gender were female; adding the 11 specimens previously identified as females, both add up to 48, the number used to size the minimum spawning mass capacity of each female.

Spawning frequency is a measure that indicates the number of masses released daily in a tank. A total number of masses were counted, as well as the number of masses whose eggs were counted and incubated and the total of eggs and larvae produced per day at the end of the incubation. The percentage of larvae hatched through incubator per day was calculated according to Vega et al. (2012), considering it as an estimation of the eggs viability and incubation process. Partial fecundity is the number of eggs per spawned mass by a female. Total fecundity of an average female is the number of eggs produced in a breeding season divided by the estimated number of females, considering a minimum of 11 and a maximum of 48. To measure eggs viability, 1 mL of egg mass was taken using a 10 mL graduated wide tip pipette, depositing it in a Petri dish, where the diameter of 10 eggs was measured through an Olympus magnifying glass (Tokyo, Japan) with a graduated lens. It was sought that eggs were transparent without dark areas, had a smooth surface with regular yolk distribution and presented an absence of parasites or microorganisms on the surface of the corium. Non-viable eggs were discarded and the viable ones were incubated. In order to estimate the number of viable eggs for incubation, 1 mL of the mass was extracted and was homogenized in a beaker, by stirring a rod in 99 mL of seawater. The number of eggs was counted 5 times under the magnifying glass, with results that average among themselves.

The equation, the Spearman's rank correlation coefficient, and the graph of relations were estimated using a database generated through the Excel 2015 program (Microsoft ©). The relations analyzed were: a) female weight *versus* volume of the masses, estimating that the greater the weight the higher the volume. The data of spawns in a tank were selected in any given month and the weights of the females were ordered from higher to lower. The masses of higher volume were assigned to the females of greater weight and the masses of lesser volumes to the females of lower weight, b) female weight *versus* number of eggs, d) volume of the mass *versus* number of eggs per 1 mL of mass.

G. chilensis is a partial spawner which can release up to two masses of eggs per day in a long reproductive season, averaging 621 ± 458 masses per season. Considering that a minimum of 11 and a maximum of 48 females exist, in the first case the average of masses/female/season is of 56 ± 42 (min 18-max 101), and in the second case is of 13 ± 10 (min 4-max 23). Three spawning seasons (I, II, III) of broodstock in captivity were identified. Season I was from September 2007 to June 2008, season II from July 2008 to May 2009 and season III from June to December 2009. Over the three seasons, 42,331,734 eggs coming from 1,864 masses were produced in total, with an average of 311 \pm 86 masses per month (min 229-max 417). The average partial fecundity of the volume of a mass was $2,563 \pm 1,172$ mL (min 200-max 6,400) and $82,186 \pm$ 44,976 eggs (min 2,000-max 284,958). The average fecundity were 1,282,780 ± 271,300 eggs/female/ season (min 997,645-max 1,537,719) in the first case, and $293,970 \pm 62,173$ (min 228,627-max 352,394) in the second case.

Directly proportional relations between the weight of the female, the volume of the mass and the numbers of eggs were identified, in which as the female weight increases, the volume of the mass and the number of eggs increase too, with high variability in the volume of the masses and the numbers of eggs for the same weight (r = 0.70 and 0.60, Fig. 1).

The relation between the volume of the mass and the number of eggs shows that as the volume increases, the number of eggs increases too, although with a low correlation (r = 0.54) due to the high variability in the number of eggs per mass (Table 1).

The total masses production at the Research Centre over the 27 months of observation and harvesting was of 2,290 masses of low viability, because only 7.0% of these (166 masses) corresponding to 15,330,517 eggs were incubated, of which only 52% one-day larvae were obtained (7,914,324; Table 2), with an average of 50,483 \pm 42,169 one-day larvae per mass.

Furthermore, an inversely proportional relationship between the number of eggs/mL/mass and the volume of the mass was identified; the greater the volume of the mass the lesser the number of eggs/mL/mass, with a high variability in the number of eggs expressed in the correlation coefficient r = 0.43 (Table 1).

An inversely proportional relation was observed between the temperature (x) and the number of eggs spawned (y), with an exponential type curve and a correlation coefficient r = 0.94 (n = 358, $y = 7E+11e^{-0.941x}$), reaching the maximum number of spawned eggs at 11-12°C (Fig. 2). The total eggs considered for this relation was 30,739,117 eggs, with an average of $85,863 \pm$ 46,524; a minimum of 2,000 and a maximum of 249,900. The same ratio was calculated using as a 'y' variable: a) the number of masses or b) the volumes of the masses, generating the following results: a) correlation coefficient r = 0.97 (n = 1,923, y = 6E+06e^{-0.788x}), the average of spawned masses was 275 ± 321 , with a minimum of 10 and a maximum of 790; b) correlation coefficient r =0.97 (n = 1,923, y = 8E+10e^{-0.914x}), the total spawned volume was of 5,120,170 mL, with an average of 2,663 \pm 1,182 mL, and a minimum of 200 and maximum of 6,400 mL.

The temperature (x) and the number of one-day larvae (y) showed an inversely proportional relationship, with a negative exponential curve (n = 143, y = $5E+10e^{-0.856x}$, correlation coefficient r = 0.88), where the maximum viability, expressed in the number of oneday larvae was obtained at 12°C, like the maximum numbers of eggs. The total number of considered larvae was of 7,508,388 ± 42,017; with a minimum of 1,162 and a maximum of 187,500.

The results obtained from 621 average masses per season for an estimation of between 11 to 48 females, confirm that *G. chilensis* is a partial spawner, as Chong *et al.* (2006) had indicated, and like *G. blacodes* and *Gadus morhua* (Paredes & Bravo, 2005; FAO, 2008). In captivity, it is important to identify and separate each female to have a specific knowledge of the volume of the mass and the number of eggs, in order to comfirm the release of more than two masses per day and the average of 13 to 56 masses/female/season. *G. chilensis* is a fish of high fecundity with an estimated average of

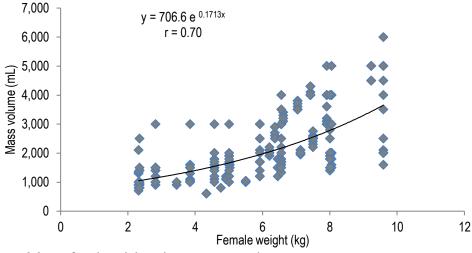


Figure 1. Genypterus chilensis female weight ratio versus mass volume.

Table 1. Summary of statistic ratios between productive parameters. r: Spearman correlation coefficient.

Ratio x - y	Equation	r	n
Female weight-mass volume	$y = 706.6 e^{0.1713x}$	0.70	18
Mass volnr. eggs	$y = -0.0009x^2 + 25.492x + 22,868$	0.54	529
Female weight-nr. eggs/mass	$y = 2831x^2 - 22201x + 98,816$	0.60	96
Mass volnr. eggs/mL/mass	$y = 1E - 06x^2 - 0.0133x + 58.013$	0.43	529

Table 2. Total masses production, eggs and one-day larvae of Genypterus chilensis in CIMARQ.

Masses	Volume (L)	Masses	Masses (%)	Eggs	One day larvae	Hatching (%)
Produced	5,862	2,290	100	-	-	-
Counted	1,408	534	23	43,962,174	-	-
Incubated	456	166	7	15,330,517	7,914,324	52

293,970 to 1,282,780 eggs/female/season, depending on the actual number of females at the Centre, like other marine fish such as *D. labrax*, *G. blacodes*, *G. morhua* and *P. adspersus* (Büke, 2002; Paredes & Bravo, 2005; FAO, 2008; Bushnell *et al.*, 2010; Rideout & Morgan, 2010; Silva & Oliva, 2010). *G. chilensis* has a positive ratio between fecundity and size. However, it comes with a high variability, even for the same size, with correlation coefficients fluctuating between 0.4 and 0.7. This variability is also reflected in species such as *G. blacodes* and *P. adspersus*. *G. blacodes* has a low total weight (TW) - or total length (TL) (r = 0.67 and 0.58) and total fecundity (TF) linear correlation (TF = 73.94 × TW - 139,356; TF = 16,003 × TL - 1,321,311 (Paredes & Bravo, 2005; Silva & Oliva, 2010).

Fishes showed sensibility to the quality parameters of water, especially to temperature, where this range is narrow and specific in each species as it was demonstrated by this study which shows that the volume of the masses (mL) and the number of eggs and spawned masses was higher between 11 and 12°C. For example, between 13 and 16°C the maximum number of eggs were obtained in *D. labrax* y *P. adspersus* as well as the maximum viability in *P. adspersus* and *P. microps*, spawning *P. adspersus* between 12.7 and 19.7°C (Büke, 2002; Silva & Oliva, 2010 fide Silva, 1996).

The percentage of fertilization and the viability of eggs at the hatching time are two parameters of quality that allow mapping out the individuals required for fish-farming. The viability of the masses of eggs in *G. chilensis* was low (7%) in comparison to that of other species such as *P. adspersus* and *P. microps*, which produce between 30 and 50% of alive floating eggs before fertilization (Silva, 2001; Silva & Oliva, 2010). The percentage of fertilization of the eggs of *G. chilensis* was not determined in this study, however, Vega *et al.* (2012) estimated that it can be as high as 80 to 87%, although in cultivated marine species there is a high variability (Table 3). It is also important to consider that there are variations in the percentage of fertilization of *G. morhua*, whose first spawning has the

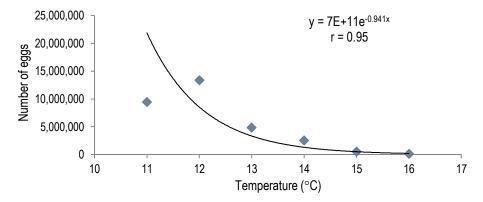


Figure 2. The ratio between temperature versus the number of eggs spawned by Genypterus chilensis.

Table 3. Fertilization rate of marine farmed fish.

Species	Fertilization rate	Hatching rate	Author
Genypterus chilensis	80 - 87		Vega et al. (2012)
Paralichthys adspersus	0 - 100		Silva & Oliva (2010)
Dicentrarchus labrax	40 - 89	74	Devauchelle & Coves (1988); Büke (2002)
Paralichthys microps	0 - 100	30 - 90	Silva (2001)
Sparus aurata	90 - 95		Moretti et al. (1999); Ouellet et al. (2001)
Gadus morhua	19 - 99		Palakovich & Kaufman (2009)

lowest value (20%), which subsequently increases for the second (84%) and third spawning (99%) (Palakovich & Kaufman, 2009).

Even though *G. chilensis* showed a low viability of the masses of eggs (7%), of the 15,330,517 eggs that were incubated, a 52% of one-day larvae survived, being these results consistent with the survivals of other marine fish like *D. labrax* (40%; Büke, 2002), *G. morhua* (35%; Ouellet *et al.*, 2001) and *P. adspersus* (30-90%; Silva & Oliva, 2010). The temperature, the number of eggs and the number of one-day larvae exhibit an inversely proportional relation, where the maximum viability was obtained at 12°C, as well as the maximum amount of eggs. These results are consistent with those of other marine species that have narrow and specific optimum temperature ranges for each species, as pointed out before.

G. chilensis produces numerous masses of eggs, of which only a small percentage reaches incubation, as well as it occurs in other marine fish. The increase of viability may be improved by taming the broodstock as well as by improving their nutrition, fertilization strategies, estimation of the quality of eggs and the incubation system.

ACKNOWLEDGMENTS

This work was supported by the FONDEF Project D06I 1024 "Development of technologies for the production of red cusk-eel fingerlings (*Genypterus chilensis*)".

REFERENCES

- Büke, E. 2002. Sea bass (*Dicentrarchus labrax* L., 1781) seed production. Turk. J. Fish. Aquat. Sci., 2: 61-70.
- Bushnell, M.E., J.T. Claisse & C.W. Laidley. 2010. Lunar and seasonal patterns in fecundity of an indeterminate, multi-spawning surgeonfish, the yellow tang *Zebrazoma flavescens*. J. Fish Biol., 76: 1343-1361. doi: 10.11 11/j.1095-8649.2010.02569.x
- Chong, J. & P. González. 2009. Ciclo reproductivo y talla media de madurez del congrio colorado, *Genypterus chilensis* (Guichenot, 1881) en el litoral de Talcahuano, Chile. Rev. Biol. Mar. Oceanogr., 44(1): 257-262.
- Chong, J., K. Sepúlveda & M. Ibáñez. 2006. Variación temporal en la dieta del congrio colorado, *Genypterus chilensis* (Guichenot, 1881) frente al litoral de Talcahuano, Chile (36°32'S, 36°45'S). Rev. Biol. Mar. Oceanogr., 41(2): 195-202.
- Food and Agriculture Organization (FAO). 2008. Cultured Aquatic Species Information Programme. *Gadus morhua*. Text by Hakon Ottera. In: FAO Fisheries and Aquaculture Department [online]. Rome. Updated January 1 2004. http://www.fao.org/ fishery/culturedspecies/Gadus_morhua/en. Reviewed: 30 June 2016.
- Kong, I., R. Meléndez & G. Henríquez. 1988. Los peces Ophidiformes de aguas profundas entre Arica e isla Mocha. Estud. Oceanol., 7: 1-15.

- Machinandiarena, L., M.F. Villarino & G.J. Macchi. 1998.
 Descripción del estadio de desove del abadejo manchado *Genypterus blacodes* (Schneider, 1801) (Pisces, Ophidiidae) en el Mar Argentino. Bol. Inst. Esp. Oceanogr., 14(1-2): 49-55.
- Olivar, M.P. & A. Sabatés. 1989. Early life history and spawning of *Genypterus capensis* (Smith, 1849) in the southern Benguela system. S. Afr. J. Mar. Sci., 8(1): 163-181. doi: 10.2989/02577618909504559.
- Ouellet, P., Y. Lambert & I. Berube. 2001. Cod egg characteristics and viability in relation to low temperature and maternal nutritional condition. ICES J. Mar. Sci., 58: 672-686. doi: 10.1006/jmsc.2001. 1065.
- Palakovich, J. & L. Kaufman. 2009. Estimating the importance of maternal age, size, and spawning experience to the recruitment of Atlantic cod (*Gadus morhua*). Biol. Conserv., 142: 477-487.
- Paredes, F. & R. Bravo. 2005. Reproductive cycle, size at first maturation and fecundity in the golden ling, *Genypterus blacodes*, in Chile. New Zeal. J. Mar. Freshw. Res., 39: 1085-1096.
- Rideout, R.M. & M.J. Morgan. 2010. Relationships between maternal body size, condition and potential fecundity of four north-west Atlantic demersal fishes.
 J. Fish Biol., 76: 1379-1395. doi: 1095-8649.2010. 02570.x.
- Servicio Nacional de Pesca (SERNAPESCA). 2016. Anuario estadístico de pesca 2014. Series. Servicio Nacional de Pesca. Ministerio de Economía, Fomento y Turismo. Valparaíso. [http://www.sernapesca.cl/ index.php?option=com_remository&Itemid=246&fun c=startdown&id=26183]. Reviewed: 27 October 2017.

Received: 8 June 2016; Accepted: 2 January 2018

- Silva, A. 2001. Advance in the culture research off smalleye flounder, *Paralichthys microps*, and Chilean flounder, *P. adspersus*, in Chile. J. Appl. Aquacult., 11(1-2): 147-164.
- Silva, A. & M. Oliva. 2010. Revisión sobre aspectos biológicos y de cultivo del lenguado chileno (*Paralichthys adspersus*). Lat. Am. J. Aquat. Res., 38(3): 377-386.
- Tascheri, R., J. Sateler, J. Merino, E. Díaz, V. Ojeda & M. Montecinos. 2003. Estudio biológico-pesquero del congrio colorado, congrio negro y congrio dorado en la zona centro sur. Fondo de investigación Pesquera, Informe Final FIP 2001-15: 300 pp.
- Universidad Andrés Bello (UNAB). 2012. Proyecto congrio colorado (*Genypterus chilensis*). Antecedentes del recurso y proyecto. [http://proyecto congrio colorado.unab.cl/]. Reviewed: 30 March 2016.
- Vega, R., M. Pradenas, J.M. Estrada, D. Ramírez, I. Valdebenito, A. Mardones, P. Dantagnan, D. Alfaro, F. Encina & C. Pichara. 2012. Evaluación y comparación de dos sistemas de incubación de huevos de *Genypterus chilensis* (Guichenot, 1848). Lat. Am. J. Aquat. Res., 40(1): 187-200.