Heritabilities and genetic correlation of shell thickness and shell length growth in a mussel, *Mytilus chilensis* (Bivalvia:Mytilidae)

Ricardo Guiñez[1], Jorge E Toro[2], Sebastián Krapivka[3], Angélica C Alcapán[2] & Pablo A Oyarzún[2]

[1]Instituto de Ciencias Naturales Alexander von Humboldt, Facultad de Ciencias del Mar y Recursos Biológicos, Universidad de Antofagasta, Antofagasta, Chile

[2]Instituto de Ciencias Marinas y Limnológicas (ICML), Universidad Austral de Chile, Valdivia, Chile

[3]Departamento de Antropología, Facultad de Ciencias Sociales, Universidad de Chile, Santiago, Chile

Correspondence: J E Toro, Instituto de Ciencias Marinas y Limnológicas (ICML), Universidad Austral de Chile, Valdivia, Chile. E-mail: jtoro@uach.cl

Abstract

Shell thickness in mollusks is generally considered adaptive because of their effects on fitness. However, little is known about the genetic basis of shell thickness. This is important, because the response to selection and the subsequent adaptive microevolution of a trait, such as thickness is only possible when that trait exhibits additive genetic variation. Here, we estimated the narrow-sense heritability (h^2: ratio between additive genetic variance and phenotypic variance) for the traits 'shell thickness' and 'shell length growth' in a 34-month-old cohort of the mussel *Mytilus chilensis* obtained by using a half-full sib design and grown in the field. Also, phenotypic and genetic correlations were estimated between both traits. We found that h^2 showed significant values for shell thickness (0.294 ± 0.194) and length (0.731 ± 0.379). The phenotypic correlation between both traits was positive and significant; however, the genetic correlation between these traits was not. These results suggest both traits can evolve adaptively by selection, but because these traits did not show genetic correlation, it is possible that selection pressure affecting one trait may not affect the other.

Keywords: heritability, shell thickness, mussels, Mytilidae, Chile, Southeast Pacific, genetic covariances

Introduction

Dissecting phenotypic variation into its genetic and plastic components represents a major challenge that permits not only a better understanding of the proximate mechanisms underlying morphological diversity, but also has potentially broader implications for evolutionary theory (McCairns & Bernatchez 2012). One way to demonstrate the interaction between genetic and environmental effects on morphological traits has been the implementation of common garden experiments and reciprocal transplants (e.g. Prada, Schizas & Yoshioka 2008; Brönmark, Lakowitz & Hollander 2011; Hice, Duffy, Munch & Conover 2012). But, even when these studies can reveal the plastic nature of the phenotype; the genetic component of it often remains implicit (McCairns & Bernatchez 2012). Without evidence for heritable variation underlying any focal trait, subsequent inference regarding its adaptive potential will remain suspect.

The narrow-sense heritability (h^2) is an important parameter of a quantitative trait because allows us to estimate the proportion of the total variance which is heritable (additive genetic variation) (Falconer & Mackay 1996; Roff 1997; Lynch & Walsh 1998). A trait can evolve by selection, either natural or artificial, when there is presence of additive genetic variation in the trait of interest (Roff 1997). The h^2 is not only fundamental when surveying the potential for genetic improvement of

a trait with a putative commercial value (Falconer & Mackay 1996), but also is important in evolutionary terms, because its value can indicate the potential for adaptive microevolutionary changes (Roff 1997), and its closeness with fitness (Mousseau & Roff 1987). Genetic correlations are also important parameters for a quantitative adaptive trait, if selection acts on only a single trait; other traits, which can be genetically correlated, may also evolve (e.g. Lande 1979).

The analysis of shell morphology and body size variation in bivalve mollusks plays an important role in many fields such as, taxonomy, genetics, selection, evolution, functional anatomy and also in fisheries management and genetic improvement of mollusks (Signorelli, Márquez & Pastorino 2013). In gastropod and bivalves mollusks, the intra- and inter-specific variation in shell morphology (i.e. shell shape and thickness) could be due to phenotypic plasticity and/or adaptive traits (e.g. Stanley 1970; Boulding & Hay 1993; Parsons 1997; Beadman, Caldow, Kaiser & Willows 2003; Ubukata 2003; Hulsey, Hendrickson & Garcia de Leon 2005; Nagarajan, Lea & Goss-Custard 2006; Signorelli *et al.* 2013; Chen, Wu, Chen & Liu 2015). Among the shell characteristics, the shell thickness and the valve length are the most influential factors for shell strength (Hamilton, Nudds & Neate 1999; Nagarajan *et al.* 2006; Lombardi, Chon, Lee, Lane & Paynter 2013). A positive correlation has been shown to exist between mussel length and force required to crush the mussels (Hamilton *et al.* 1999). The shell thickness can protect individuals against physical stressors (Seed & Richardson 1999; Pascoal, Carvalho, Creer, Mendo & Hughes 2012) such as the destructive effects of intense wave action (Guiñez 1996; Steffani & Branch 2003), as well as a protection against predation (Reimer & Tedengren 1996; Leonard, Bertness & Yund 1999; Smith & Jennings 2000; Caro & Castilla 2004), or can provide a mechanical support for the effects of density and packing/aggregation in mussel beds or matrices (Guiñez & Castilla 1999; Guiñez, Petraitis & Castilla 2005; Briones, Rivadeneira, Fernández & Guiñez 2014). However, despite the ecological, evolutionary and productive importance of the shell thickness in bivalves, according to our knowledge, there is no a single report in the literature on the estimation of its heritability (h^2). In a critical search and analysis of the literature, we found two studies that measured the heritability of

the shell thickness in mollusks, Chaves-Campos, Coghill, Al-Salamah, DeWitt and Johnson (2012) in the snail *Mexipyrgus churinceanus*, and Jin, Bai, Fu, Zhang and Li (2012) in the bivalve *Hyriopsis cumingii*. However, Jin *et al.* (2012) properly measured the body maximum latero-lateral length axe (see Fig. 1 in Guiñez & Castilla 1999), also defined as shell width by Wada (1986), and shell breadth by Wang, Du, Lü and Liu (2010), but they did not measure the shell thickness as traditionally defined (the valve thick). Much more focus has been done with estimations of heritability values for body size, and all of its surrogates: mass, length and other linear dimensions (e.g. Newkirk 1980; Gjedrem 1983; Collet, Boudry, Thebault, Heurtebise, Morand & Gerard 1999; Jones, Bates, Innes & Thompson 1996; Toro, Alcapán, Vergara & Ojeda 2004b; Alcapán, Néspolo & Toro 2007; Jin *et al.* 2012).

The aim of this research was to estimate the heritability (h^2) and the phenotypic and genetic correlation of shell thickness and maximum length, as a surrogate of body size, in a 34-month-old cohort of *M. chilensis*, using a half/full-sib design. In *M. chilensis* larger shell thickness have been reported within wild individuals in contrast to suspended cultivated populations (Valladares, Manriquez & Suarez-Isla 2010); and it has been suggested that wild mussel bed populations may face higher predation pressures or other environmental stress factors than cultured mussels. However, shell thickness in *M. chilensis* could be an adaptive or a plastic phenotypic trait.

The Chilean blue mussel, *Mytilus chilensis* (Hupe) is a common intertidal and subtidal bivalve
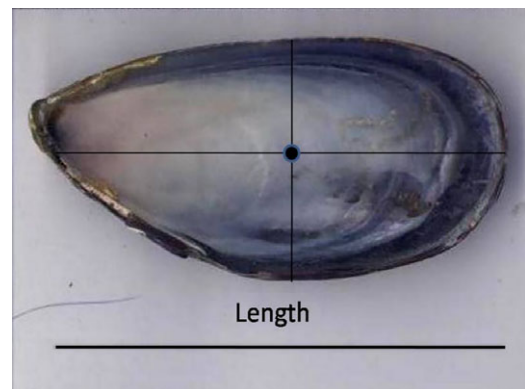

Figure 1 Shell length dimension (mm) and location of the point where shell thickness (mm) using an outside point caliper was measured in each valve. [Colour figure can be viewed at wileyonlinelibrary.com].

species of Central-South Chile (Krapivka, Toro, Alcapan, Astorga, Presa, Perez & Guinez 2007), they can be found forming dense beds in sedimentary areas of the inland coast of the north Patagonic archipelagos of the Chilean coast (ca. 40–43°S), and their role as ecosystem engineers has been also studied (Duarte, Jaramillo, Contreras & Figueroa 2006; Buschbaum, Dittmann, Hong, Hwang, Strasser, Thiel, Valdivia, Yoon & Reise 2009; Aldea & Rosenfeld 2011). This mussel is also under intensive aquaculture production in southern Chile and these cultured mussels are mainly exported to Spain, France, and the United States (SERNAPESCA 2014). This mussel industry (over 350 000 t year^{-1}) until now relies only on natural spat fall (Sernapesca 2014). However, if a breeding programme could develop faster growing mussels, with specific properties, farmers would readily use hatchery-produced spat. Hence, the study of quantitative genetics of size-related traits and other properties, such as the shell thickness, in the blue mussel are important either because of basic (i.e. ecological-evolutionary knowledge) and/or practical (i.e. animal production and wild bed management) purposes. Studies performed in this species have reported narrow-sense heritability values on larvae and adult size-related traits, generating the quantitative genetics basis for applied production programmes (Toro 1995; Toro & Paredes 1996; Toro, Alcapán, Ojeda & Vergara 2004a; Toro, Alcapán, Vergara *et al.* 2004b; Alcapán *et al.* 2007).

Material and methods

Adult individuals of *Mytilus chilensis* were obtained from natural subtidal beds at Yaldad Bay (43°50′24″S, 73°30′5″W) and transferred to Hueihue (41°50′24″S, 73°30′5″W) on the East coast of Chiloé Island (Chile). Breeding was implemented as a half/full-sib design (Lynch & Walsh 1998): each male (sire) was bred with five females (dams). Spawning was induced in water baths at 19.5°C, during 2 h, male and female gametes were collected separately and breeding was performed as a half – sib configuration (Lynch & Walsh 1998): each male (= sire) was bred with five females (= dams). To accomplish this, spawning of females was induced by adding 80 mL of water with the specific sire gametes to the selected dam, and observing cellular division after 1.5 h. A total of 19 sires and 95 dams were successfully spawned.

Larvae were maintained in water at 18 ± 1°C in an open circulation system, and fed daily with micro – algae mostly composed by *Isochrysis galbana* (10^5 cells mL^{-1}) (Toro & Paredes 1996). The embryos from each family were placed into a 200 L fibre-glass tank containing 1 μm filtered and U.V. treated fresh sea water (FSW) 16 ± 1°C, at a density of 100 individuals per ml. After 24 h (D-stage larvae) the density was adjusted to 5 larvae per millilitre. A high cell concentration of the micro algae *Isochrysis galbana* (Parke 1949) and *Chaetoceros gracilis* (Schuett) were used 100 000 cells mL^{-1} as food concentration (Toro & Paredes 1996). Every day, the water in each tank was passed through a 45-micron 'nitex' screen to retain the larvae. Each tank was rinsed with fresh water followed by seawater. The larvae were then resuspended in FSW and algal food was added daily at the desired cell concentration. Juveniles from each family were settled on netlon mesh and maintained in culture tanks until a mean size of 3 ± 1 mm. Then, families distributed in labelled pearl nets were transferred to the field (Quetalmahue; 41°51′S, 73°55′W) in long lines (suspended culture), until 10 month of age. At this age, they all individuals within each family were tagged and then separated into three groups, maintaining the sib composition (i.e. each half–sib family was divided by three). One group was randomly distributed in pearl nets and maintained in Quetalmahue, the second group was moved to Putemún (42°25′50″S, 73°43′45″W) and the third to Hueihue (41°51′S, 73°30′W). After 34 months, a storm affected severely the families from Quetalmahue and Hueihue, but we were able to rescue almost all the families with the exception of three full-sib families lost from the locality of Putemún. So, in this study, we used only the individuals from Putemún with 12 half-sib families and 60 full-sib families. Each mussel was measured for shell thickness and length at both left and right valves, and both values (left and right) were averaged for statistical analyses. The shell thickness was measured as the valve's thickness, at the mid-point of each valve using an outside point caliper (± 0.0005 mm), and the length as the maximum antero-posterior value with a caliper (± 0.005 mm) (Fig. 1).

All the linear-mixed models were run with PROC MIXED (SAS Institute Inc., Cary, NC, USA) using restricted maximum-likelihood method (REML), which was used instead of least squares

methods because our design was unbalanced (Littell, Milliken, Stroup & Wolfinger 1996). A full/half-sib design was conducted, using a nested where the trait of each offspring of each dam was the dependent variable. The first level was sires and the second level, nested within sires was dams. Therefore, genetic variances/covariances and heritabilities estimates included sire and dam nested within sire as random effects. The causal variance components were calculated using the estimated variance components (among sire: σ_s^2; among dam: σ_d^2; among progeny: σ_p^2) (Falconer & Mackay 1996). The additive genetic variance (V_A) was calculated as four times the among-sire variance ($V_A = 4\,\sigma_s^2$), the maternal/common environmental variances as, $V_M = \sigma_d^2 - \sigma_s^2$, and the residual variance V_R as the among progeny variance (= within full-sib family variance; $V_R = \sigma_p^2$). And the phenotypic variance (V_P), was estimated as $V_P = V_A + V_M + V_R$. The narrow-sense heritability (h^2) was estimated as $h^2 = V_A/V_P = 4\sigma_s^2/V_p$ (Roff 1997). Significance of the respective genetic component of each model was assessed by comparing the full model with a reduced model lacking the additive genetic component using a log-likelihood ratio test (Saxton 2004). The standard error of heritability values was computed following Roff (1997). In addition, we used WOMBAT to estimate heritability, the successor of DFREML – an animal model- implemented by Meyer (2007) for mixed-model analyses using REML. The phenotypic correlation between both traits was determined using Pearson product-moment correlation using procedure CORR (SAS Institute). Additive genetic correlation, r_A, was estimated as, $r = COV_A/\sqrt{V_{A1}\,V_{A2}}$ where COV_A is the covariance between the additive genetic values for the two traits (shell thickness and shell length) and V_{A1} and V_{A2} are the additive genetic variances for each trait. Significance of the genetic covariance was assessed using a likelihood ratio test by comparing the likelihood of the model containing the genetic covariance component with the reduced model in which the genetic covariance was fixed at 0 (Saxton 2004).

Results

Shell thickness showed a higher coefficient of variation (CV) than shell length (Table 1). Nonetheless, both CV were high, indicating that they have high phenotypic variances. Additive genetic variances (sire component) were significant for both traits (Table 2) but maternal and common environmental effects represented by the dam (sire) component were significant only for shell length ($P < 0.0001$, Table 2). The estimation of heritability (h^2) values of shell thickness and length were significantly different from 0 and similar for both PROC MIXED and the animal model WOMBAT ($P < 0.0135$, Table 3) and highest for shell length than for thickness (Table 3). Phenotypic correlation was positive and significantly different from 0 ($P < 0.0001$), but genetic correlation was positive but not statistically significant ($P = 0.147$, Table 3).

Discussion

Our experiment revealed high individual variability for shell thickness (CV = 35.59) and length growth (CV = 11.41) in *M. chilensis*. However, this variability could be explained by the presence of phenotype plasticity or adaptive processes (Pigliucci 2001). Inducible defenses – a type of phenotypic plasticity – (Bourdeau, Butlin, Bronmarck, Edgell, Hoverman & Hollander 2015) are a pervasive feature of species found on littoral assemblages (Leonard *et al.* 1999; Bourdeau 2011; Pascoal *et al.* 2012), certainly an increase in shell thickness is a good mechanism of defense to reduce the efficiency of their predators (Leonard *et al.* 1999; Smith & Jennings 2000; Caro & Castilla 2004; Freeman 2007; Pascoal *et al.* 2012). In other cases, the differences in shell thickness between populations can be explained by adapta-

Table 1 Descriptive statistics of shell length (mm) and shell thickness (mm) of *Mytilus chilensis*. Mean ± 1 standard error. CV is the coefficient of variation

Trait	Sire	Dam	N	Min–Max	Mean, mm	CV
Length	12	60	345	56.51–99.56	73.34 ± 0.45	11.41
Thickness	12	60	351	0.44–2.66	1.07 ± 0.02	35.59

Table 2 Estimation of genetic covariance (± standard error) using linear-mixed model analysis of shell thickness (mm) and shell length (mm) with PROC MIXED. In round parenthesis, the variance components and in brackets the probability of the log-likelihood ratio test

Trait	Sire	Dam (sire)	Residual
Shell thickness	0.0113 ± 0.0075 (0.0452 ± 0.0298) [0.0135]	0.0080 ± 0.0064 (0.0033 ± 0.0106) [0.1473]	0.1277 ± 0.0106 (0.1051 ± 0.0183) [<0.0001]
Shell length	23.1657 ± 12.0184 (92.6628 ± 48.0736) [<0.0001]	19.3262 ± 5.0794 (3.8395 ± 13.4295) [<0.0001]	30.3123 ± 2.5458 (30.3123 ± 24.1706) [<0.0001]

Table 3 Heritabilities (bold), genetic correlation (upper triangle) and phenotypic correlation (lower triangle) (±standard error) for shell thickness and length in *M. chilensis*. In brackets probability of the log-likelihood ratio test. Heritabilities in bold were estimated by linear mixed model using PROC MIXED, and heritabilities in *italic* were estimated using an animal model with a linear-mixed model via restricted maximum likelihood (REML) as implemented in WOMBAT. In round parenthesis, the probability of the Pearson correlation coefficient

	Shell thickness	Shell length
Shell thickness	**0.294 ± 0.194** [0.0135] *0.241 ± 0.158* [<0.0001]	0.5482 ± 0.300 [0.147]
Shell length	0.255 ± 0.370 (<0.0001)	**0.731 ± 0.379** [<0.0001] *0.931 ± 0.367* [<0.0001]

tions to the presence or absence of predators by selective pressures, as for example, the case of *Mytilus* from the North Sea and the Baltic Sea (Kautsky, Johannesson & Tedengren 1990). Also, it has been suggested that inducible defenses (e.g. Shell thickness) are adaptive responses of prey to environments in which predation pressure varies spatially or temporally (Leonard *et al.* 1999; Smith & Jennings 2000; Freeman 2007; Lowen, Innes & Thompson 2013; Bourdeau *et al.* 2015). Despite these facts, to our knowledge there was no estimation of the additive genetic variance of shell thickness itself and of their plasticity in bivalves. If there is a significant heritability ($h^2 > 0$) then adaptive microevolutionary changes of the shell thickness can occur. Indeed, it can be predicted that shell thickness and shell length growth in *M. chilensis* can evolve responding to natural or artificial selection, as both heritabilities are greater than zero.

With respect to the growth in shell length, this is the third report of narrow-sense heritability for

this trait. The first estimate of half-sib heritability ranged between 0.38 ± 0.33 and 0.84 ± 0.45 in larval and spat stages (Toro, Alcapán, Vergara *et al.* 2004b), the second one ranged between 0.010 ± 0.146 and 0.520 ± 0.100 at ages between 12 and 22 months including three localities (Hueihue, Putemún and Quetalmahue) (Alcapán *et al.* 2007) and in this work at 34 months ranged between 0.731 ± 0.379 and 0.931 ± 0.367. The heritability values estimated in this study are larger than those described in the literature for the same species; among some of the causes (change in heritability values along the life animal span or environmental causes), the measure of right and left valves, and using the average of both in the analysis could be an explanation. This procedure reduces significantly the residual error variance error (V_R) in the analysis (Briones & Guiñez 2005; Lajus, Katolikova, Strelkov & Hummel 2015), and therefore decreasing the phenotypic variation (denominator) thus increasing the additive variance.

In addition, we found evidence of a significant positive phenotypic correlation between shell thickness and shell length growth. It is well established that the shell thickness of mussels has a phenotypic correlation with their length (Nagarajan, Lea & Goss-Custard 2002; Nagarajan *et al.* 2006), and this might also be taken as an initial indication of a genetic correlations between these traits. However, the data presented here showed that both traits were not genetically correlated, suggesting that these traits can be affected by a different set of genes. This indicates that selection acting on one trait (e.g. shell thickness) may not necessary affect the other trait (e.g. length shell growth).

As stated before, the additive genetic variation is not only important for improvement purposes in

important economic traits, but also to study the microevolution of these traits under different scenarios. For example, thicker shells found in natural populations, in contrast to thinner shells in cultivated populations of *M. chilensis* (Valladares *et al.* 2010) can be the results of selection due to differential predation pressures acting at both kinds of populations. In fact, *M. chilensis* shows several identified predators (Gordillo & Amuchastegui 1998; Gordillo 2001; Navarro, Leiva, Gallardo & Varela 2002; Andrade & Ríos 2007; Gordillo & Archuby 2012), so the next step, is to test if the variation in selective pressures (e.g. by predators) influences Darwinian fitness (Pigliucci 2001). This study is contributing to this issue; however, more field studies are needed, to understand the adaptation under selective pressures in *Mytilus chilensis*.

Acknowledgments

References

Alcapán A.C., Néspolo R.F. & Toro J.E. (2007) Heritability of body size in the Chilean blue mussel (*Mytilus chilensis* Hupe 1854): effects of environment and ageing. *Aquaculture Research* **38**, 313–320.

Aldea C. & Rosenfeld S. (2011) Intertidal macromolluscs from the rocky substrata of Buque Quemado Beach, Strait of Magellan, southern Chile. *Revista de Biología Marina y Oceanografía* **46**, 115–124.

Andrade C. & Ríos C. (2007) Estudio experimental de los habitos tróficos de *Trophon geversianus* (Pallas 1774) (Gastropoda: Muricidae): selección y manipulación de presas. *Anales Instituto de la Patagonia (Chile)* **35**, 45–53.

Beadman H.A., Caldow R.W.G., Kaiser M.J. & Willows R.I. (2003) How to toughen up your mussels: using mussel shell morphological plasticity to reduce predation losses. *Marine Biology* **142**, 487–494.

Boulding E.G. & Hay T.K. (1993) Quantitative genetics of shell form of an intertidal snail: constraints on short-term response to selection. *Evolution* **47**, 576–592.

Bourdeau P.E. (2011) Constitutive and inducible defensive traits in co-occirring marine snails distributed across a vertical rocky intertidal gradient. *Functional Ecology* **25**, 177–185.

Bourdeau P.E., Butlin R.K., Bronmarck C., Edgell T.C., Hoverman J.T. & Hollander J. (2015) What can aqua-

tic gastropods tell us about phenotypic plasticity? A review and meta-analysis. *Heredity* **115**, 312–321.

Briones C & Guiñez R. (2005) Asimetria bilateral de la forma de las valvas y posición especial en matrices del chorito *Perumytilus purpuratus* (Lamarck, 1819) (Bivalvia: Mytilidae). *Revista Chilena de Historia Natural* **78**, 3–14.

Briones C., Rivadeneira M.M., Fernández M. & Guiñez R. (2014) Geographical variation of shell thickness in the mussel *Perumytilus purpuratus* along the southeast Pacific coast. *The Biological Bulletin* **227**, 221–231.

Brönmark C., Lakowitz T. & Hollander J. (2011) Predator-induced morphological plasticity across local populations of a freshwater snail. *PLoS ONE* **6**, e21773.

Buschbaum C., Dittmann S., Hong J.S., Hwang I.S., Strasser M., Thiel M., Valdivia N., Yoon S.P. & Reise K. (2009) Mytilid mussels: global habitat engineers in coastal sediments. *Helgoland Marine Research* **63**, 47–58.

Caro A.U. & Castilla J.C. (2004) Predator-inducible defences and local intrapopulation variability of the intertidal mussel *Semimytilus algosus* in central Chile. *Marine Ecology Pregress Series* **276**, 115–123.

Chaves-Campos J., Coghill L.M., Al-Salamah M.A., DeWitt T.J. & Johnson S.G. (2012) Field heritabilities and lack of correlation of snail shell form and anti-predator function estimated using Bayesian and maximum likelihood methods. *Evolutionary Ecology Research* **14**, 743–755.

Chen Y.J., Wu J.Y., Chen C.T.A. & Liu L.L. (2015) Effecs of low-pH stress on shell traitsof the dove snail, *Anachis misera*, inhabiting shallow-vent environments off Kueishan Islet, Taiwan. *Biogeosciences* **12**, 2631–2639.

Collet B., Boudry P., Thebault A., Heurtebise S., Morand B. & Gerard A. (1999) Relationship between pre- and post-metamorphic growth in the Pacific oyster *Crassostrea gigas* (Thunberg). *Aquaculture* **175**, 215–226.

Duarte C., Jaramillo E., Contreras H. & Figueroa L. (2006) Community structure of the macroinfauna in the sediments below an intertidal mussel bed (*Mytilus chilensis* (Hupe) of southern Chile. *Revista Chilena de Historia Natural* **79**, 353–368.

Falconer D.S. & Mackay T.F.C. (1996) *Introduction to Quantitative Genetics*. Longman Group Ltd, Harlow, Essex, England.

Freeman A.S. (2007) Specificity of induced defenses in *Mytilus edulis* and asymmetrical predator deterrence. *Marine Ecology Progress Series* **334**, 145–153.

Gjedrem T. (1983) Genetic variation in quantitative traits and selective breeding in fish and shellfish. *Aquaculture* **33**, 51–72.

Gordillo S. (2001) Predation signs by *Acanthina* Fischer von Waldheim, 1807 (Gasteropoda: Muricidae) on Bivalvia. *Ameghiniana* **38**, 55–60.

Gordillo S. & Amuchastegui S.N. (1998) Predation strategies of shell-boring gastropods *Trophon Geversianus* (Pallas) (Muricoidea: Trophonidae). *Malacologia* **39**, 83–91.

Gordillo S. & Archuby F. (2012) Predation by drilling gastropods and asteroids upon mussels in rocky shallow shores of southernmost South America: paleontological implications. *Acta Palaeontologica Polonica* **57**, 633–646.

Guiñez R. (1996) Dinámica poblacional del chorito maico, *Perumytilus purpuratus* (Lamarck 1819) (Bivalvia: Mytilidae), en gradientes de exposición al oleaje. Unpublished D. Phil., Thesis, Pontificia Universidad Católica de Chile, Santiago, Chile.

Guiñez R. & Castilla J.C. (1999) A tridimensional self-thinning model for multilayered intertidal mussels. *American Naturalist* **154**, 341–357.

Guiñez R., Petraitis P.S. & Castilla J.C. (2005) Layering, the effective density of mussels and mass-density boundary curves. *Oikos* **110**, 186–190.

Hamilton D.J., Nudds T.D. & Neate J. (1999) Size-selective predation of blue mussels (*Mytilus edulis*) by common eiders (*Somateria mollissima*) under controlled field conditions. *The Auk* **116**, 403–416.

Hice L.A., Duffy T.A., Munch S.B. & Conover D.O. (2012) Spatial scale and divergent patterns of variation in adapted traits in the ocean. *Ecology Letters* **15**, 568–575.

Hulsey C.D., Hendrickson D.A. & Garcia de Leon F.J. (2005) Thophic morphology, feeding performance and prey use in the polymorphic fish *Herichthys minckleyi*. *Evolutionary Ecology Research* **7**, 303–324.

Jin W., Bai Z.Y., Fu L.L., Zhang G.F. & Li J.L. (2012) Genetic analysis of early growth traits of the triangle shell mussel, *Hyriopsis cumingii*, as an insight for potential genetic improvement to pearl quality and yield. *Aquaculture International* **20**, 927–933.

Jones R., Bates J.A., Innes D.J. & Thompson R.J. (1996) Quantitative genetic analysis of growth in larval scallops (*Placopecten magellanicus*). *Marine Biology* **124**, 671–677.

Kautsky N., Johannesson K. & Tedengren M. (1990) Genotypic and phenotypic differences between Baltic and North Sea populations of *Mytilus edulis* evaluated through reciprocal transplantations. I Growth and morphology. *Marine Ecology Progress Series* **59**, 203–2010.

Krapivka S., Toro J.E., Alcapan A.C., Astorga M., Presa P., Perez M. & Guinez R. (2007) Shell-shape variation along the latitudinal range of the Chilean blue mussel *Mytilus chilensis* (Hupe 1854). *Aquaculture Research* **38**, 1770–1777.

Lajus D., Katolikova M., Strelkov P. & Hummel H. (2015) Fluctuating and directional asymmetry of the blue mussel (*Mytilus edulis*): improving methods of morphological analysis to explore species performance at the northern border of its range. *Symmetry* **7**, 488–514.

Lande R. (1979) Quantitative genetic analysis of multivariate evolution, applied to brain: body size allometry. *Evolution* **33**, 402–416.

Leonard G.H., Bertness M.D. & Yund P.O. (1999) Crab predation, waterborne cues, and inducible defenses in the blue mussel, *Mytilus edulis*. *Ecology* **80**, 1–14.

Littell R.C., Milliken G.A., Stroup W.W. & Wolfinger R.D. (1996) *SAS System for Mixed Models*. SAS Institute, Cary, NC, USA.

Lombardi S.A., Chon G.D., Lee J.J., Lane H.A. & Paynter K.T. (2013) Shell hardness and compressive strength of the eastern oyster, *Crassostrea virginica*, and the asian oyster, *Crassostrea ariakensis*. *The Biological Bulletin* **225**, 175–183.

Lowen J.B., Innes D.J. & Thompson R.J. (2013) Predator-induced defenses differ between sympatric *Mytilus edulis* and *M. trossulus*. *Marine Ecology Progress Series* **475**, 135–143.

Lynch M. & Walsh B. (1998) *Genetics and Analysis of Quantitative Traits*. Sinauer Associates, Inc., Sunderland, MA, USA.

McCairns R.J.S. & Bernatchez L. (2012) Plasticity and heritability of morphological variation within and between parapatric stickleback demes. *Journal of Evolutionary Biology* **25**, 1097–1112.

Meyer K. (2007) WOMBAT—A tool for mixed model analyses in quantitative genetics by restricted maximum likelihood (REML). *Journal of Zhejiang University Science B* **8**, 815–821.

Mousseau T.A. & Roff D.A. (1987) Natural selection and the heritability of fitness components. *Heredity*, **59**, Part 2, 181–197.

Nagarajan R., Lea S.E.G. & Goss-Custard J.D. (2002) Reevaluation of patterns of mussel (*Mytilus edulis*) selection by European Oystercatchers (*Haematopus ostralegus*). *Canadian Journal of Zoology* **80**, 846–853.

Nagarajan R., Lea S.E.G. & Goss-Custard J.D. (2006) Seasonal variations in mussel, *Mytilus edulis* L. shell thickness and strength and their ecological implications. *Journal of Experimental Marine Biology and Ecology* **339**, 241–250.

Navarro J.M., Leiva G.E., Gallardo C.S. & Varela C. (2002) Influence of diet and temperature on physiological energetics of *Chorus giganteus* (Gastropoda: Muricidae) during reproductive conditioning. *New Zealand Journal of Marine and Freshwater Research* **36**, 321–332.

Newkirk G.F. (1980) Review of the genetics and the potential for selective breeding of commercially important bivalves. *Aquaculture* **19**, 209.

Parsons K.E. (1997) Contrasting patterns of heritable geographic variation in shell morphology and growth potential in the marine gastropod *Bembicium vittatum*: evidence from field experiments. *Evolution* **51**, 784–796.

Pascoal S., Carvalho G., Creer S., Mendo S. & Hughes R. (2012) Plastic and heritable variation in shell thickness of the intertidal gastropod *Nucella lapillus* associated with risks of crab predation and wave action, and sexual maturation. *PLoS ONE* **7**, e52134.

Pigliucci M. (2001) *Phenotypic Plasticity: Beyond Nature and Nurture.* Johns Hopkins Press, Baltimore, MD, USA.

Prada C., Schizas N. & Yoshioka P. (2008) Phenotypic plasticity or speciation? A case from a clonal marine organism. *BMC Evolutionary Biology* **8**, 1–19.

Reimer O. & Tedengren M. (1996) Phenotypical improvement of morphological defenses in the mussel *Mytilus edulis* induced by exposure to the predator Asterias rubens. *Oikos* **75**, 383–390.

Roff D.A. (1997) *Evolutionary Quantitative Genetics.* Chapman & Hall, New York, USA.

Saxton A. (2004) *Genetic Analysis of Complex Traits Using SAS.* SAS Institute, Cary, NC, USA.

Seed R. & Richardson C.A. (1999) Evolutionary traits in *Perna viridis* (Linnaeus) and *Septifer virgatus* (Wiegmann) (Bivalvia: Mytilidae). *Journal of Experimental Marine Biology and Ecology* **239**, 273–287.

Sernapesca. (2014) *Anuarios Estadísticos de Pesca.* Servicio Nacional de Pesca, Chile.

Signorelli J.H., Márquez F. & Pastorino G. (2013) Phenotypic variation of south-western Atlantic clam *Mactra isabelleana* (Bivalvia: Mactridae). *Journal of the Marine Biology Association of United Kingdom* **93**, 511–517.

Smith L.D. & Jennings J.A. (2000) Induced defensive responses by the bivalve *Mytilus edulis* to predators with different attack modes. *Marine Biology* **136**, 461–469.

Stanley S.M. (1970) Relation of shell form to life habitats in the Bivalvia (Mollusca). *GSA Memoirs* **125**, 1–296.

Steffani C.N. & Branch G.M. (2003) Growth rate, condition, and shell shape of *Mytilus galloprovincialis*: responses to wave exposure. *Marine Ecology Progress Series* **246**, 197–209.

Toro J.E. (1995) Response to artificial selection and realized heritability estimates for live weight and shell length for Chilean native oyster (*Ostrea chilensis* Philippi 1845). *Aquaculture* **137**, 271–272.

Toro J.E. & Paredes L.I. (1996) Heritability estimates of larval shell length in the Chilean blue mussel *Mytilus chilensis*, under different food densities. *Aquatic Living Resources* **9**, 347–350.

Toro J.E., Alcapán A.C., Ojeda J.A. & Vergara A.M. (2004a) Selection response for growth rate (shell height and live weight) in the Chilean blue mussel (*Mytilus chilensis* Hupe 1854). *Journal of Shellfish Research* **23**, 753–757.

Toro J.E., Alcapán A.C., Vergara A.M. & Ojeda J.A. (2004b) Heritability estimates of larval and spat shell height in the Chilean blue mussel (*Mytilus chilensis* Hupe 1854) produced under controlled laboratory conditions. *Aquaculture Research* **35**, 56–61.

Ubukata T. (2003) A morphometric study on morphological plasticity of shell form in crevice-dwelling Pterioida (Bivalvia). *Biological Journal of the Linnean Society* **79**, 285–297.

Valladares A., Manriquez G. & Suarez-Isla B.A. (2010) Shell shape variation in populations of *Mytilus chilensis* (Hupe 1854) from southern Chile: a geometric morphometric approach. *Marine Biology* **157**, 2731–2738.

Wada K.T. (1986) Genetic selection for shell traits in the Japanese pearl oyster, *Pinctada fucata martensii*. *Aquaculture* **57**, 171–176.

Wang H., Du X., Lü W. & Liu Z. (2010) Estimating the heritability for growth-related traits in the pearl oyster, *Pinctada fucata martensii* (Dunker). *Aquaculture Research* **42**, 57–64.