

# Marine Biology Research

**Marine Biology Research** 

ISSN: 1745-1000 (Print) 1745-1019 (Online) Journal homepage: http://www.tandfonline.com/loi/smar20

### Age estimation methods in the marine gastropod Buccinanops globulosus comparing shell marks and opercula growth rings

Verena Bökenhans, Gregorio Bigatti & Andres Averbuj

To cite this article: Verena Bökenhans, Gregorio Bigatti & Andres Averbuj (2016): Age estimation methods in the marine gastropod Buccinanops globulosus comparing shell marks and opercula growth rings, Marine Biology Research, DOI: 10.1080/17451000.2016.1209526

To link to this article: http://dx.doi.org/10.1080/17451000.2016.1209526



Published online: 09 Sep 2016.



Submit your article to this journal 🗹



View related articles



則 🛛 View Crossmark data 🗹

Full Terms & Conditions of access and use can be found at http://www.tandfonline.com/action/journalInformation?journalCode=smar20

#### **ORIGINAL ARTICLE**



## Age estimation methods in the marine gastropod *Buccinanops globulosus* comparing shell marks and opercula growth rings

Verena Bökenhans<sup>a,b</sup>, Gregorio Bigatti<sup>a,b</sup> and Andres Averbuj<sup>a</sup>

<sup>a</sup>LARBIM-IBIOMAR, Centro Nacional Patagónico – CONICET, Puerto Madryn, Argentina; <sup>b</sup>Facultad de Ciencias Naturales, Universidad Nacional de la Patagonia San Juan Bosco (UNPSJB), Puerto Madryn, Argentina

#### ABSTRACT

Age determination in gastropods is an essential tool for understanding the main aspects of population dynamics that allow for the formation of fisheries policies. Small-scale fisheries of the nassariid gastropod *Buccinanops globulosus* from northern Patagonia are not yet regulated, although it is locally consumed while an incipient commercialization is taking place. The aim of this study was to show the suitability of opercula readings for age estimation in *B. globulosus*, compared with shell marks, previously validated by the analysis of the stable isotope of oxygen. The individual age estimated by the opercula rings reading method did not match the estimation made by the shell marks reading method. Stable isotope analyses confirmed that shell marks were deposited annually. Proper age estimation for *B. globulosus*, based on recognizable external shell marks, reached up to 9 years.

#### **ARTICLE HISTORY** Received 3 March 2016

Accepted 27 June 2016

**RESPONSIBLE EDITOR** Francesc Maynou

KEYWORDS Edible gastropod; fisheries; individual growth; Nassariidae; stable oxygen isotopes

#### Introduction

Fisheries sustainability depends on population parameter studies as a first step for correct management (Botsford et al. 1997). Small-scale fisheries, in particular benthic invertebrate fisheries, have played an important role in the development of new fishery management principles and tools (Leiva & Castilla 2002). Many marine organisms that possess hard structures such as gastropods with calcareous shells and statoliths or proteic opercula, exhibit conspicuous growth band patterns that may allow an estimation of age, if this banding pattern is a consequence of a periodic event.

Previous studies on marine gastropods, including caenogastropods of the South Atlantic region, suggested age determination to be an essential tool for the understanding of the main aspects of population dynamics (Kideys 1996; Ilano et al. 2004; Bigatti et al. 2007). There are several methods that are commonly used to estimate age in gastropods; among them statolith ring formation has been proved to be a consistent method (Richardson et al. 2005a, 2005b; Galante-Oliveira et al. 2015). Another reliable age estimation technique based on the shell markings has been used recently in many gastropod species. This method requires the validation by oxygen stable isotope analysis of the seasonality of the calcareous deposition that produces the shell marks (Epstein et al. 1953; Schöne et al. 2003; Giménez et al. 2004; Bigatti et al. 2007; Arrighetti et al. 2012). This validation allows for the determination of the time interval between two consecutive discernible marks (changes in colour and aspect) in the gastropod shell, and provides a direct assessment of individual age (Santarelli & Gros 1985).

At the same time, the opercular rings of many gastropod species have been used to estimate their ages; however, proper validation of their seasonality is usually absent (Kideys 1996; Ilano et al. 2004; Narvarte et al. 2008; Avaca et al. 2013).

Buccinanops globulosus (Kiener, 1834) is an edible nassariid gastropod endemic to the south-western Atlantic Ocean. It inhabits shallow coastal waters from Uruguay (34°S) to southern Argentina (48°S) (Scarabino 1977). It is consumed locally in northern Patagonia, where an annual catch of 9200 kg in 2004 (Narvarte 2006) has been reported. This species is, at present, commercially exploited and exported to the Asian market (Averbuj et al. 2014; Bigatti et al. 2015).

The aim of this study is to show the suitability of opercular readings for age estimation, compared with shell marks, previously validated by stable isotope analysis of oxygen, in the edible gastropod *B. globulosus* from Golfo Nuevo, Atlantic North Patagonia.

CONTACT Andres Averbuj 🖾 averbuj@cenpat-conicet.gob.ar 🗈 LARBIM-IBIOMAR, Centro Nacional Patagónico – CONICET, Bvd. Brown 2915, U9120ACV Puerto Madryn, Chubut, Argentina

 $<sup>\</sup>ensuremath{\mathbb{C}}$  2016 Informa UK Limited, trading as Taylor & Francis Group

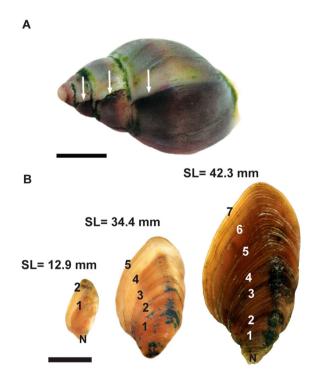
#### **Materials and methods**

#### Sampling

A total of 224 individuals of Buccinanops globulosus representing all size ranges were collected in Cerro Avanzado (42°49'S, 64°52'W), from a sandy beach in Golfo Nuevo, Atlantic North Patagonia, during April 2013. Samplings were taken using baited traps (using decomposed lamb meat) to attract the gastropods, anchored at 2 to 5 metres depth during low tide. Individuals were randomly collected from the traps. Sampled individuals represented the complete size range of the population. In the laboratory the shells were separated from the soft body and total shell length measured to the nearest 0.1 mm with a calliper. The operculum was carefully removed from the foot, cleaned, labelled (coinciding with the individual's shell) and stored in dry conditions for later analyses. The monthly average water temperature data provided by the Naval Hydrographic Service was used (SHN 2014).

#### Stable isotope analysis

Stable oxygen isotope ratio ( $\delta^{18}$ O) was used to analyse the frequency of shell mark formation. Two specimens of *Buccinanops globulosus* were used for stable isotope



**Figure 1.** A: Shell marks of *Buccinanops globulosus* (white arrows). B: Opercula showing growth rings (numbers), nucleus (N) and shell length (SL) of the specimen from which each operculum was obtained. Scale bars: A - 1 cm, B - 0.50 cm.

analysis, measuring 31.02 mm (# 1) and 37.22 mm (# 2). Each carbonate sample consisted of 10 mg of powder drilled from the shell. Samples were taken from the prismatic layer of the shell along the spiral growth ring with special attention being given to drill on each of the visible external marks and between them. Because of the small size of the shells and the large amount of carbonate samples needed for the technique, a limited number of samples could be drilled in the shell. Isotopic ratios were determined as described by Zabala et al. (2013). The results were reported as a deviation per mil ( $\delta^{18}O$  ‰) relative to the VPDB (Vienna Pee Dee Belemnite) standard and the analytical error was 0.1‰ for  $\delta^{18}O$ .

During shell deposition, the ratio of stable oxygen isotopes (<sup>18</sup>O and <sup>16</sup>O) in the biogenic of CaCO<sub>3</sub> is inversely proportional to temperature (Epstein et al. 1951). As demonstrated by Epstein & Lowenstam (1953) and Epstein et al. (1953), this relationship is almost linear between 5° and 30°C. Thus, in the study area, where the salinity is constant and a cyclical annual oscillation of temperature is present, we expect the  $\delta^{18}$ O curve to reflect the environmental seasonality (Giménez et al. 2004; Cledón et al. 2005; Bigatti et al. 2007; Arrighetti et al. 2011).

 $\delta^{18}$ O values were converted into water temperature by the palaeotemperature equation of Epstein et al. (1953), as modified by Craig (1965):

$$\begin{split} T^{^{\circ}}(C) &= 16.00 - 4.14 (\delta^{18} O_{carbonate} - \delta^{18} O_{water}) \\ &+ 0.13 (\delta^{18} O_{carbonate} - \delta^{18} O_{water})^2. \end{split}$$

Isotopic ratios of a seawater sample were also measured at INGEIS (Instituto de Geocronología y Geología Isotópica). Sea water  $\delta^{18}O_{water}$  was estimated at -0.3%.

#### Shell mark and operculum ring quantification

Shell marks (SM) and opercular growth rings (OR) were counted by two observers. If the data counted by both observers was not equal, it was discarded. A total of 132 pairs of data were obtained.

Each shell was gently smoothed with fine sandpaper, rinsed in seawater and clean dried with absorbent paper. Previous studies for caenogastropods confirmed the correspondence of internal and external marks through X-ray photography of shells (Giménez et al. 2004; Cledón et al. 2005; Bigatti et al. 2007; Arrighetti et al. 2011, 2012; Zabala et al. 2013), including a species of the same genus, *Buccinanops cochlidium* (Dillwyn, 1817) (Averbuj 2009). The SM that corresponded to maximum values of stable oxygen isotope analysis coincided with a colour change that covered the total width of the shell (straight line), perpendicular to the direction of growth (Figure 1A).

Unbroken opercula with clearly distinguishable OR were used, as explained by Kideys (1996). Growth rings are seen as dark rings between areas of lighter colour (Figure 1B). Each operculum length was measured.

The total number of SM and the number of visible OR were counted under a stereoscopic microscope.

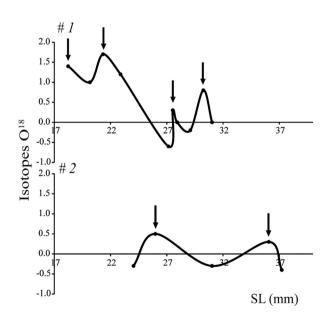
#### Statistical analysis

Data were computed as 132 pairs of SM and OR observations, one pair for each individual. The fit of the data to a linear regression analysis between SM and OR was compared with a linear model for which slope is equal to 1 and the intercept equal to 0, using maximum like-lihood methods (Hilborn & Mangel 1997). The coefficient of probability and optimization routine *solver* provided by Microsoft Excel was used for this purpose.

Additionally an age bias plot (which assesses for the consistency of age determination) and the coefficient of variation (CV) (measure of precision) were incorporated following Campana et al. (1995).

#### **Results**

The 224 animals collected ranged from 6.55 to 49.15 mm in length (SL) (mean  $\pm$  SD = 26.78  $\pm$  10.08 mm), while the opercula ranged from 4.54 to



**Figure 2.**  $\delta^{18}$ O profile plotted against corresponding shell lengths (SL) of two individuals of *Buccinanops globulosus*. Black arrows correspond to shell marks.

17.15 mm in length (10.69  $\pm$  2.99 mm). The SL was significantly related to the opercular length ( $R^2 = 0.934$ ; F = 1849.79; *P*-values < 0.0001) by the following linear regression:

$$y = 2.9109x - 0.6522.$$

#### Stable oxygen isotopes

The  $\delta^{18}$ O profile of *Buccinanops globulosus* shells reflected a seasonal water temperature regime (Figure 2). The average  $\delta^{18}$ O profiles ranged between -0.05% and 0.725%, which corresponds to a temperature range of about  $11.89^{\circ}$ C and  $14.96^{\circ}$ C. Visible SM coincided with the maximum values of  $\delta^{18}$ O (corresponding to minimum temperature), i.e. one shell growth mark was formed each year when water temperature was minimal.

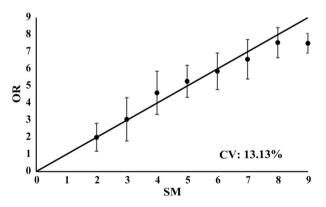
#### Shell mark and operculum ring quantification

The total number of SM and OR ranged from 1 to 9, corresponding to individuals of all sizes. The maximum age of 9 years (recorded by SM) corresponded to a shell length of 45.19 mm in total.

A significant linear regression was found between the SM and the OR (CA:  $R^2 = 0.627$ , N = 132, F = 219.30, *P*-values < 0.0001), as follows:

$$SM = 0.757 \times OR - 1.4145$$

Non-linear systematic differences in age estimation were present (Figure 3). A coefficient of variation (CV) of 13.13% was estimated. Coincidences in the pairs of observed SM and OR were found in only 25% of the cases (N = 132).



**Figure 3.** Age bias plot for the pairwise age estimation methods. Each error bar represents the 95% confidence interval about the mean age assigned by opercula rings (OR) for all specimens of a given age assigned by shell marks (SM). The 1:1 equivalence (solid line) is also indicated. A non-linear systematic difference in age estimation is present. CV: Coefficient of variation.

**Table I.** Linear regression analysis between shell marks and opercula rings. Comparison of the slope and the intercept, to a slope = 1 and intercept = 0 with the Maximum Likelihood Ratio test. CV: Critical value, df: degrees of freedom.

	CV	$\chi^2$ critic	df	P value
Slope	21.19	3.841	1	4.15E <sup>-06</sup>
Intercept	21.85	3.841	1	2.93E <sup>-06</sup>

Data did not fit significantly a linear model with a slope equal to one and intercept equal to zero (Table I).

#### Discussion

Several techniques have been used to estimate age for gastropods, using different validation methods. Counting the number of surface rings on the operculum has been widely used (Kideys 1996; Chen & Soong 2002; llano et al. 2004; Narvarte 2006; Avaca et al. 2013); however, the validation is uncertain. Richardson et al. (2005a) proposed a clear validation for the use of statolith rings as age estimators. Another reliable technique is the use of oxygen isotope analysis of different shell sections (Cledón et al. 2005; Bigatti et al. 2007; Arrighetti et al. 2011, 2012) taking into account the ratio of stable oxygen isotopes (<sup>18</sup>O and <sup>16</sup>O) in mollusc shells during the deposition of CaCO<sub>3</sub> that depends on seawater isotope composition and temperature (Brey & Mackensen 1997).

The formation of the shell marks was demonstrated to be an annual event by stable isotope analysis. Although it is difficult to accurately validate the periodicity of the growth ring formation in the operculum, the inability of the data to fit a linear model, where the slope equals 1 and the intercept equals 0, suggested that both methods do not estimate age equivalently in Buccinanops globulosus. This is also evident in the systematic differences between the two estimation methods, observed as non-linear age estimation bias. According to this, age estimated by opercula rings was overestimated in median ages and underestimated in older ages. The plot together with the CV coefficient provides a powerful and easily prepared comparison of matched pairs of age determinations (Campana et al. 1995). According to the present study, the age estimation of B. globulosus based on the number of shell marks is reliable and should be used instead of reading opercular rings. Thus, from our estimation, the maximum age in the studied population of B. globulosus was 9 years.

The shell formation process in molluscs implies building an organic membrane, composed of a gel phase (silk), a structural phase (chitin) and a matrix of macromolecules where the mineralization occurs (Falini et al. 1996; Addadi et al. 2006; Furuhashi et al. 2009, 2010). The calcium and carbonate present in the seawater are used by the organisms during crystallization (Bevelander 1952; Falini et al. 1996; Cespuglio et al. 1999; Graham & Sarikaya 2000). The isotopic ratio incorporated during crystallization varies with temperature (Epstein & Lowenstam 1953). Therefore, the isotopic ratio present in the shell of the mollusc reflects seawater temperature and provides a precise age estimation in temperate or cold regions where the temperature shows an annual oscillation (Richardson 2001), as is the case of the Golfo Nuevo in northern Patagonia (Dellatorre et al. 2012).

The first detailed study of the operculum formation was proposed by Houssay (1884). He suggested that the visible rings on the operculum outer surface are formed by successive appositions of new secreted material from the pedal fissure. These materials are proteins where the amino acids vary slightly depending on the species, associated with small amounts of carbohydrates including neutral and amino sugars (Hunt 1971, 1976). Proteins are a metabolic product (Morais et al. 2003; Ren et al. 2003; Najmudeen 2007) and their availability depends on the gastropod diet and physical condition, such as gonadic development stage, environmental factors affecting metabolism and the quantity and nutritional value of the food intake (Whyte et al. 1990, Morais et al. 2003). Thus, a direct relationship between temperature and the number of opercula rings is not expected, but the causes of deposition could not be explained by our results.

Shell marks and opercula rings are clearly visible by a thickening, and a clear change of colour, probably corresponding to a slower growth during a short period of adverse environmental conditions, as described by Vasconcelos et al. (2012). Individuals that presented a clearly recognizable mark, observed as an edge thickening, were considered to have completed a whole year. Shells that showed high levels of erosion or damage were not taken into account. Opercula with undistinguished rings caused by erosion, deformation or broken edges were also discarded.

Previous studies of *B. globulosus* (Narvarte et al. 2008; Avaca et al. 2013) attempted to demonstrate the annual formation of the opercular rings, lacking a proper comparison with a reliable validated technique. Comparative age estimating techniques have previously been used in other species to validate the operculum rings. Richardson et al. (2005a) demonstrated an annual periodicity of deposition in the statolith ring formation by corresponding it with the ratio of Mg:Ca present in the shell of *Neptunea antiqua* (Linnaeus,

1758) (Buccinidae). When comparing the number of statolith and opercular rings, there was a significant difference between the numbers obtained by both techniques, similar to that registered in our study. Santarelli & Gros (1985) compared the position of the opercular rings with one of the shell marks that corresponded to the maximum value of  $\delta^{18}$ O, by the use of sclerochronology in three individuals of *Buccinum undatum* Linnaeus, 1758. In that paper they also found that the opercular rings did not coincide with the formation of the shell marks.

Our study allows for growth modelling studies to be applied to fishery management in a much more precise and reliable way. The age estimating technique of opercular ring reading requires an effective validation of its seasonality. Proper age estimation for *B. globulosus*, based on the results obtained with stable isotope analyses, could be accomplished by counting the clearly recognizable external shell marks, for individuals up to 9 years old. However, in areas where the shells are subject to high erosion this method should be used with caution, accompanied by stable isotope analysis.

#### Acknowledgements

The authors are members of CONICET. All research work, including sampling and laboratory investigations, complies with current Argentine laws. Drs Miguel Pascual, Martin Garcia-Azorey and Juan Pablo Livore provided helpful suggestions that greatly improved the manuscript.

#### **Disclosure statement**

No potential conflict of interest was reported by the authors.

#### Funding

This work was supported by FONCyT (Fondo para la Investigación Científica y Tecnológica), including projects PICT 1232, PICT 2929, PICT 1491, PICT 1476, and CONICET (Consejo Nacional de Investigaciones Científicas y Técnicas), including project PIP 067.

#### References

- Addadi L, Joester D, Nudelman F, Weiner S. 2006. Mollusk shell formation: a source of new concepts for understanding biomineralization processes. Chemistry – A European Journal 12:980–87. doi:10.1002/chem.200500980
- Arrighetti F, Brey T, Mackensen A, Penchaszadeh PE. 2011. Age, growth and mortality in the giant snail *Adelomelon beckii* (Broderip 1836) on the Argentinean shelf. Journal of Sea Research 65:219–23. doi:10.1016/j.seares.2010.10.003

- Arrighetti F, Teso V, Brey T, Mackensen A, Penchaszadeh PE. 2012. Age and growth of *Olivancillaria deshayesiana* (Gastropoda: Olividae) in the southwestern Atlantic Ocean. Malacologia 55:163–70. doi:10.4002/040.055.0111
- Avaca MS, Narvarte M, Martín P. 2013. Age, growth and mortality in *Buccinanops globulosus* (Gastropoda: Nassariidae) from Golfo Nuevo (Argentina). Marine Biology Research 9:208–19. doi:10.1080/17451000.2012.708420
- Averbuj A. 2009. Biologia Reproductiva, Crecimiento y Dieta del Caracol Ciego Buccinanops cochlidium (Gastropoda: Nassariidae) en Golfo San José, Argentina. Doctoral Thesis. Universidad de Buenos Aires (UBA), Buenos Aires. 186 pages.
- Averbuj A, Rocha MN, Zabala S. 2014. Embryonic development and reproductive seasonality of *Buccinanops globulosus* (Nassariidae) (Kiener, 1834) in Patagonia, Argentina. Invertebrate Reproduction & Development 58:138–47. doi:10.1080/07924259.2013.875949
- Bevelander G. 1952. Calcification in molluscs. III. Intake and deposition of Ca<sup>45</sup> and P<sup>32</sup> in relation to shell formation. Biological Bulletin 102(9):9–15. doi:10.2307/1538617
- Bigatti G, Penchaszadeh PE, Cledon M. 2007. Age and growth in *Odontocymbiola magellanica* (Gastropoda: Volutidae) from Golfo Nuevo, Patagonia, Argentina. Marine Biology 150:1199–204. doi:10.1007/s00227-006-0401-6
- Bigatti G, Cumplido M, Averbuj A. 2015. Gastropods of commercial interest in the Province of Chubut. Technical Report for Mesa Técnica Zona 1 – Subsecretaría de Pesca de Chubut – Reglamentación de la Pesca de Gasterópodos. Inf. N° 30 del LAPEMAR, Laboratorio de Peces y Mariscos de Interés Comercial (CENPAT). 39 pages.
- Botsford LW, Castilla JC, Peterson CH. 1997. The management of fisheries and marine ecosystems. Science 277:509–15. doi:10.1126/science.277.5325.509
- Brey T, Mackensen A. 1997. Stable isotopes prove shell growth bands in the Antarctic bivalve *Laternula elliptica* to be formed annually. Polar Biology 17:465–68. doi:10. 1007/s003000050143
- Campana SE, Annand MC, McMillan JI. 1995. Graphical and statistical methods for determining the consistency of age determinations. Transactions of the American Fisheries Society 124:131–38. doi:10.1577/1548-8659 (1995)124<0131:GASMFD>2.3.CO;2
- Cespuglio G, Piccinetti C, Longinelli A. 1999. Oxygen and carbon isotope profiles from *Nassa mutabilis* shells (Gastropoda): accretion rates and biological behaviour. Marine Biology 135:627–34. doi:10.1007/s002270050663
- Chen M-H, Soong K. 2002. Estimation of age in the sex-changing, coral-inhabiting snail *Coralliophila violacea* from the growth striae on opercula and a mark-recapture experiment. Marine Biology 140:337–42. doi:10.1007/ s002270100700
- Cledón M, Brey T, Penchaszadeh P, Arntz W. 2005. Individual growth and somatic production in *Adelomelon brasiliana* (Gastropoda; Volutidae) off Argentina. Marine Biology 147:447–52. doi:10.1007/s00227-005-1589-6
- Craig H. 1965. The measurement of oxygen isotope paleotemperatures. In: Tongiorgi E, editor. Stable Isotopes in Oceanographic Studies and Paleotemperatures. Spoleto, Italy: Consiglio Nazionale delle Ricerche (CNR), p 161–82.
- Dellatorre FG, Pisoni JP, Barón PJ, Rivas AL. 2012. Tide and wind forced nearshore dynamics in Nuevo Gulf (Northern

Patagonia, Argentina): potential implications for crossshore transport. Journal of Marine Systems 96-97:82–89. doi:10.1016/j.jmarsys.2012.02.004

- Epstein S, Lowenstam HA. 1953. Temperature-shell-growth relations of recent and interglacial Pleistocene shoal-water biota from Bermuda. The Journal of Geology 61:424–38. doi:10.1086/626110
- Epstein S, Buchsbaum R, Lowenstam H, Urey HC. 1951. Carbonate-water isotopic temperature scale. Geological Society of America Bulletin 62:417–26. doi:10.1130/0016-7606(1951)62[417:CITS]2.0.CO;2
- Epstein S, Buchsbaum R, Lowenstam HA, Urey HC. 1953. Revised carbonate-water isotopic temperature scale. Geological Society of America Bulletin 64:1315–26. doi:10.1130/0016-7606(1953)64[1315:RCITS]2.0.CO;2
- Falini G, Albeck S, Weiner S, Addadi L. 1996. Control of aragonite or calcite polymorphism by mollusk shell macromolecules. Science 271:67–69. doi:10.1126/science.271.5245.67
- Furuhashi T, Schwarzinger C, Miksik I, Smrz M, Beran A. 2009. Molluscan shell evolution with review of shell calcification hypothesis. Comparative Biochemistry and Physiology B 154:351–71. doi:10.1016/j.cbpb.2009.07.011
- Furuhashi T, Miksik I, Smrz M, Germann B, Nebija D, Lachmann B, et al. 2010. Comparison of aragonitic molluscan shell proteins. Comparative Biochemistry and Physiology B 155:195–200. doi:10.1016/j.cbpb.2009.11.007
- Galante-Oliveira S, Marçal R, Espadilha F, Sá M, Abell R, Machado J, Barroso CM. 2015. Detection of periodic Sr  $Ca^{-1}$  cycles along gastropod statoliths allows the accurate estimation of age. Marine Biology 162:1473–83. doi:10. 1007/s00227-015-2684-y
- Giménez J, Brey T, Mackensen A, Penchaszadeh P. 2004. Age, growth, and mortality of the prosobranch *Zidona dufresnei* (Donovan, 1823) in the Mar del Plata area, south-western Atlantic Ocean. Marine Biology 145:707–12.
- Graham T, Sarikaya M. 2000. Growth dynamics of red abalone shell: a biomimetic model. Materials Science and Engineering C 11:145–53. doi:10.1016/S0928-4931(00) 00202-2
- Hilborn R, Mangel M. 1997. The Ecological Detective: Confronting Models with Data. Princeton, NJ: Princeton University Press. 336 pages.
- Houssay M. 1884. On the operculum of the gasteropoda. Journal of Natural History 13:304–06. doi:10.1080/ 00222938409459240
- Hunt S. 1971. Comparison of three extracellular structural proteins in the gastropod mollusc *Buccinum undatum* L., the periostracum, egg capsule and operculum. Comparative Biochemistry and Physiology B 40:37–40. doi:10.1016/ 0305-0491(71)90059-9
- Hunt S. 1976. The gastropod operculum: a comparative study of the composition of gastropod opercular proteins. Journal of Molluscan Studies 42:251–60.
- Ilano AS, Ito A, Fujinaga K, Nakao S. 2004. Age determination of *Buccinum isaotakii* (Gastropoda: Buccinidae) from the growth striae on operculum and growth under laboratory conditions. Aquaculture 242:181–95. doi:10.1016/j. aquaculture.2004.03.028
- Kideys A. 1996. Determination of age and growth of *Buccinum* undatum L. (Gastropoda) off Douglas, Isle of Man. Helgoländer Meeresuntersuchungen 50:353–68. doi:10. 1007/BF02367109

- Leiva GE, Castilla JC. 2002. A review of the world marine gastropod fishery: evolution of catches, management and the Chilean experience. Reviews in Fish Biology and Fisheries 11:283–300. doi:10.1023/A:1021368216294
- Morais S, Boaventura D, Narciso L, Ré P, Hawkins SJ. 2003. Gonad development and fatty acid composition of *Patella depressa* Pennant (Gastropoda: Prosobranchia) populations with different patterns of spatial distribution, in exposed and sheltered sites. Journal of Experimental Marine Biology and Ecology 294:61–80. doi:10.1016/ S0022-0981(03)00258-2
- Najmudeen TM. 2007. Variation in biochemical composition during gonad maturation of the tropical abalone *Haliotis varia* Linnaeus 1758 (Vetigastropoda: Haliotidae). Marine Biology Research 3:454–61. doi:10.1080/17451000701696 252
- Narvarte MA. 2006. Biology and fishery of the whelk *Buccinanops globulosum* (Kiener, 1834) in northern coastal waters of the San Matías Gulf (Patagonia, Argentina). Fisheries Research 77:131–37. doi:10.1016/j.fishres.2005.11.006
- Narvarte M, Willers V, Avaca MS, Echave ME. 2008. Population structure of the snail *Buccinanops globulosum* (Prosobranchia, Nassariidae) in San Matías Gulf, Patagonia Argentina: isolated enclaves? Journal of Sea Research 60:144–50. doi:10.1016/j.seares.2008.04.005
- Ren JS, Marsden ID, Ross AH, Schiel DR. 2003. Seasonal variation in the reproductive activity and biochemical composition of the Pacific oyster (*Crassostrea gigas*) from the Marlborough Sounds, New Zealand. New Zealand Journal of Marine and Freshwater Research 37:171–82. doi:10. 1080/00288330.2003.9517155
- Richardson C. 2001. Molluscs as archives of environmental change. Oceanography and Marine Biology Annual Review 39:103–64.
- Richardson C, Saurel C, Barroso C, Thain J. 2005a. Evaluation of the age of the red whelk *Neptunea antiqua* using statoliths, opercula and element ratios in the shell. Journal of Experimental Marine Biology and Ecology 325:55–64. doi:10.1016/j.jembe.2005.04.024
- Richardson C, Kingsley-Smith P, Seed R, Chatzinikolaou E. 2005b. Age and growth of the naticid gastropod *Polinices pulchellus* (Gastropoda: Naticidae) based on length frequency analysis and statolith growth rings. Marine Biology 148:319–26. doi:10.1007/s00227-005-0072-8
- Santarelli L, Gros P. 1985. Détermination de l'âge et de la croissance de *Buccinum undatum* L. (Gasteropoda: Prosobranchia) à l'aide des isotopes stables de la coquille et de l'ornementation operculaire. Oceanologica Acta 8:221–29.
- Scarabino V. 1977. Moluscos del Golfo San Matías (Provincia de Rio Negro, Republica Argentina). Inventario y claves para su identificacion. Comunicaciones de la Sociedad Malacológica del Uruguay 4:177–285.
- Schöne B, Tanabe K, Dettman D, Sato S. 2003. Environmental controls on shell growth rates and  $\delta^{18}$ O of the shallow-marine bivalve mollusk *Phacosoma japonicum* in Japan. Marine Biology 142:473–85.
- SHN. 2014. Servicio de Hidrografía Naval. http://www.hidro. gob.ar/ (accessed 2014).
- Vasconcelos P, Gharsallah IH, Moura P, Zamouri-Langar N, Gaamour N, Missaoui H, et al. 2012. Appraisal of the usefulness of operculum growth marks for ageing

*Hexaplex trunculus* (Gastropoda: Muricidae): comparison between surface striae and adventitious layers. Marine Biology Research 8:141–53. doi:10.1080/17451000.2011. 616896

Whyte J, Englar J, Carswell B. 1990. Biochemical composition and energy reserves in *Crassostrea gigas* exposed to

different levels of nutrition. Aquaculture 90:157-72. doi:10.1016/0044-8486(90)90338-N

Zabala S, Penchaszadeh PE, Panarello HO, Brogger MI, Bigatti G. 2013. Life history parameters in the edible snail *Adelomelon ancilla* from Patagonian waters. Malacologia 56:215–29. doi:10.4002/040.056.0212