Journal of FISH BIOLOGY

fsbi An International Society for Fish Biology

Journal of Fish Biology (2016) **88**, 1413–1429
doi:10.1111/jfb.12917, available online at wileyonlinelibrary.com

Reproductive biology of the southern thorny skate *Amblyraja doellojuradoi* (Chondrichthyes, Rajidae)

G. Delpiani*

Laboratorio de Biotaxonomía Morfológica y Molecular de Peces (BIMOPE), Instituto de Investigaciones Marinas y Costeras (IIMyC), Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Universidad Nacional de Mar del Plata, Funes 3350, Mar del Plata, Argentina

(Received 19 November 2015, Accepted 20 January 2016)

The total lengths (L_T) of 193 males (209–556 mm) and 130 females (275–515 mm) of *Amblyraja doellojuradoi*, a commercial by-catch species on the Argentinean continental shelf, which are increasingly retained, were analysed. No sexual dimorphism was observed in the L_T at which 50% of individuals were sexually mature; males matured at 448 mm and females at 411 mm, *c.* 80 and 82% of maximum L_T. The hepato-somatic index was similar among sexes, but significantly different between maturity stages, being lower in mature than immature specimens. Males had no seasonal difference in the hepato-somatic index and females had the lowest index in autumn. The gonado-somatic index was lower in males than in females and significantly higher in mature than immature specimens of both sexes. Males had the highest index in autumn and females had no seasonal difference. Collectively, these results would indicate that *A. doellojuradoi* breeds in autumn.

Key words: gonado-somatic index; hepato-somatic index; reproduction; south-west Atlantic Ocean.

INTRODUCTION

Chondrichthyans display a wide range of reproductive strategies (Musick & Ellis, 2005). This diversity is often associated with the environmental conditions inhabited by the skate (Rajidae) species, with deep-water skates more likely to deposit eggs year-round (García *et al.*, 2008), whereas shallow-water skates are more likely to have seasonal reproduction (Braccini & Chiaramonte, 2002; Mabragaña *et al.*, 2002, 2014) as deeper waters are subject to less environmental variability (Hamlett & Koob, 1999; Ruocco *et al.*, 2006; Scenna & Díaz de Astarloa, 2014). In the south-west Atlantic Ocean, reproductive modes and strategies are associated with the zoogeographic provinces (Colonello, 2009). Thus, in the Argentine Province, oviparous species such as *Sympterygia bonapartii* Müller & Henle, 1841 (Mabragaña *et al.*, 2002), *Sympterygia acuta* Garman 1877 (Mabragaña *et al.*, 2014), *Rioraja agassizii* (Müller & Henle 1841) (Oddone *et al.*, 2007; Colonello, 2009), *Psammobatis rudis* Günther 1870 and *Psammobatis normani* McEachran 1983 (Mabragaña & Cousseau, 2004) have annual reproductive cycles with seasonal peaks. In the Magellanic Province,

Tel.: +54 223 4753150; email: gabriela.delpiani@gmail.com

species of the genus *Bathyraja* Ishiyama 1958 show no peaks of reproductive activity (Ruocco *et al.*, 2006; Scenna, 2011), a fact that has been linked to the stability in the physical conditions of their deeper environment (Colonello, 2009).

Skates have a relatively long juvenile stage, concentrating their energy into reproductive activities and once mature their growth rate decreases. As a result, many species reach maturity only when they reach between 75 and 90% of their maximum total length (L_T) (Dulvy *et al.*, 2000; Ebert, 2005). Then, they have late maturity, high juvenile survivorship, slow body growth and long life span (Winemiller & Rose, 1992; Camhi *et a*l., 1998; Matta & Gunderson, 2007; Saglam & Ak, 2012); therefore, they cannot sustain high levels of fishing pressure (McPhie & Campana, 2009). Skates are captured around the world by commercial and recreational fishing, mainly as by-catch in fisheries targeting other species (Walker, 1998; Stevens *et al.*, 2000; Tamini *et al.*, 2006; Enever *et al.*, 2009). As the fisheries expand, secondary species [rays (Myliobatidiformes) and other elasmobranchs] may be gaining greater commercial importance and may become target species of directed fisheries. In Argentina, the decline of commercial teleost stocks has contributed to the increased landings of skates since 1994 (Lasta *et al.*, 2001), which increased from 1000 t in 1994 to 15 000 t in 2001 (Massa *et al.*, 2004).

The southern thorny skate *Amblyraja doellojuradoi* (Pozzi 1935) is a by-catch species and is increasingly being retained. Studies on the reproductive biology of skates are therefore essential to assess the population's status and propose fishery management strategies. The objective of the present study was to examine the reproduction cycle of *A. doellojuradoi* collected on the Argentinean continental shelf.

MATERIALS AND METHODS

SAMPLING

The specimens were collected from research cruises carried out by the Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP) ($n = 210$) and from commercial vessels ($n = 113$) between 2005 and 2012 in the south-west Atlantic Ocean between 36° and 50° S, from 75 to 414 m depth (Fig. 1). Available location data for specimens obtained by commercial vessels were fishing quadrants. Analyses were conducted by season (autumn, $n = 170$; winter, $n = 29$; spring, $n = 112$; summer, $n = 29$), since no samples were obtained every month, thus precluding a monthly assessment of reproductive variables.

During laboratory processing, all animals were sexed, measured from the tip of the snout to the tip of the tail (L_T) and disc width (W_D), total mass (M), liver mass (M_L) and gonad mass (M_G) were recorded.

In females, the oviducal gland width (W_{OD}), uterus width (W_U), follicle mass and diameter and egg capsule presence were measured and weighed. In females, the freeze and thaw process compromised follicle integrity, preventing accurate measurements of ovary mass (M_O), number and diameter of the ovarian follicles. This is reflected in the variation in the number of specimens that are considered for each analysis.

In males, the clasper length (L_C, measured from the distal end of the cloaca to the distal end of the clasper), the number of alar thorns, testes mass (M_T) and degree of epididymal coiling was measured and recorded.

BIOLOGICAL DATA

The size distributions of both sexes by season were plotted at intervals of 50 mm. The relationships between L_T and M were determined and the null hypothesis of no differences between

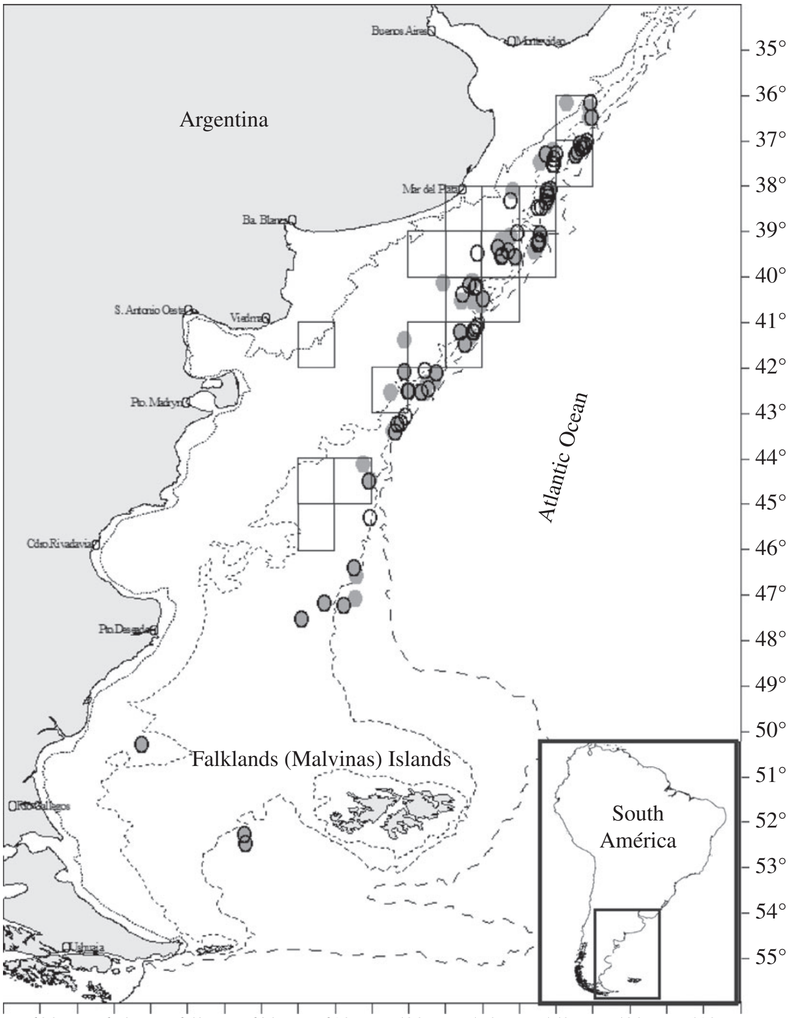

FIG. 1. Map of the study area, showing the position of fishing sets and quadrant (□) where *Amblyraja doellojuradoi* were captured. ○, males; ●, female.

slopes of both sexes was evaluated using ANCOVA (Crawley, 2005). To evaluate differences in sex ratio, a χ^2 test was used (Zar, 1984) for the entire sample.

MATURATION

Maturity stages of *A. doellojuradoi* were determined by macroscopic observation of the reproductive organs (Mabragaña *et al.*, 2002; Colonello *et al.*, 2007*a*).

For both sexes, the proportion of mature individuals in 20 mm L_T intervals was calculated and a logistic curve was fitted to the data using a maximum-likelihood approach in order to estimate the size at which 50% of individuals ($L_{T50\%}$) were sexually mature (Roa *et al.*, 1999). In order to examine if differences between the curves of both sexes exist, a likelihood ratio test was performed (Aubone & Wohler, 2000).

REPRODUCTIVE ANALYSIS

The relationships between L_T and M_T, the L_C and N_{AT} in males were analysed; and in females M_O, W_{OD} and W_U. Differences between ovary mean mass, number and diameter of the ovarian follicles of right and left gonads was assessed by a Wilcoxon test (Crawley, 2005). It was assessed whether the number and diameter of ovarian follicles correlates with female L_T. The frequency distribution of the maximum diameter of the ovarian follicles of mature females was plotted.

The hepato-somatic ($I_H = 100\, M_L\, M^{-1}$) and gonado-somatic ($I_G = 100\, M_G\, M^{-1}$) indices were calculated and compared between sexes by Mann–Whitney *U*-tests. These indices were also calculated for maturity stages and compared using a Kruskal–Wallis test. If any differences were found, a Tukey's test was performed to identify which stages were different. In females, the relationships among maximum oocyte diameter and the I_H and I_G were analysed by Spearman correlation coefficient. In males, the relationship between I_H and I_G was analysed to determine the seasonality of the reproductive cycle.

In addition, seasonal variations in the reproductive organs of *A. doellojuradoi* were evaluated to assess the reproductive cycle. The variation of I_H, I_G, W_{OD}, maximum oocyte diameter and number of oocytes of mature specimens was evaluated in relation to seasons using a Kruskal–Wallis test.

RESULTS

BIOLOGICAL DATA

Three hundred and twenty-three samples were collected in this study of which 193 were males and 130 were females. The L_T ranges observed were 209–556 mm for males and 275–515 mm for females. The main difference between seasons was the higher numbers captured in autumn ($n = 170$) and the lower number in summer ($n = 12$). The largest number of specimens measured between 450 and 499 mm L_T and sizes between 500 and 550 mm were principally males [Fig. 2(a), (b)]. The relationship between L_T and M differs significantly between the sexes (ANCOVA, $F_{130,190} = 1063{\cdot}58$, $P < 0{\cdot}001$) [Fig. 2(c)].

The male:female sex ratio of 1·48:1·00 was not significantly different from 1:1 ($G = 6{\cdot}14$, d.f. = 1, $P > 0{\cdot}05$). In autumn, the sex ratio was significantly different from 1:1 (males:females; 1·86:1·00; $G = 7{\cdot}53$, d.f. = 3, $P < 0{\cdot}05$). Within mature animals, mature males were more prominent in the samples than mature females (1·51:1; $G = 5{\cdot}64$, d.f. = 1, $P < 0{\cdot}05$) and was due to the significant differences in autumn (males:females; 2·11:1·00; $G = 8{\cdot}43$, d.f. = 3, $P < 0{\cdot}05$). The number of immature (M1 and M2) and mature males (M3) differed significantly (immature:mature; 1·00:4·07; $G = 35{\cdot}46$, d.f. = 1, $P < 0{\cdot}001$) and this ratio was maintained during the sampling period, with more mature than immature males in autumn (immature:mature; 1·00:4·68; $G = 22{\cdot}69$, d.f. = 3, $P < 0{\cdot}05$) and spring (immature:mature; 1·00:6·75; $G = 17{\cdot}06$, d.f. = 3, $P < 0{\cdot}05$). The number of immature females (F1 and F2) differed significantly from the number of mature females (F3 and F4) (immature:mature; 1·00:3·64; $G = 21{\cdot}06$, d.f. = 1, $P < 0{\cdot}001$) with a more mature than immature females in autumn (immature:mature; 1·00:2·62; $G = 5{\cdot}83$, d.f. = 3, $P < 0{\cdot}05$) and spring (immature:mature; 1·00:5·25; $G = 11{\cdot}56$, d.f. = 3, $P < 0{\cdot}05$).

MATURATION

The scale of sexual maturity in males was evaluated in terms of development of the sperm ducts, testes and the calcification of the clasper; and in females considering the uterus, ovaries, oviducal glands and the presence of egg capsules (Table I).

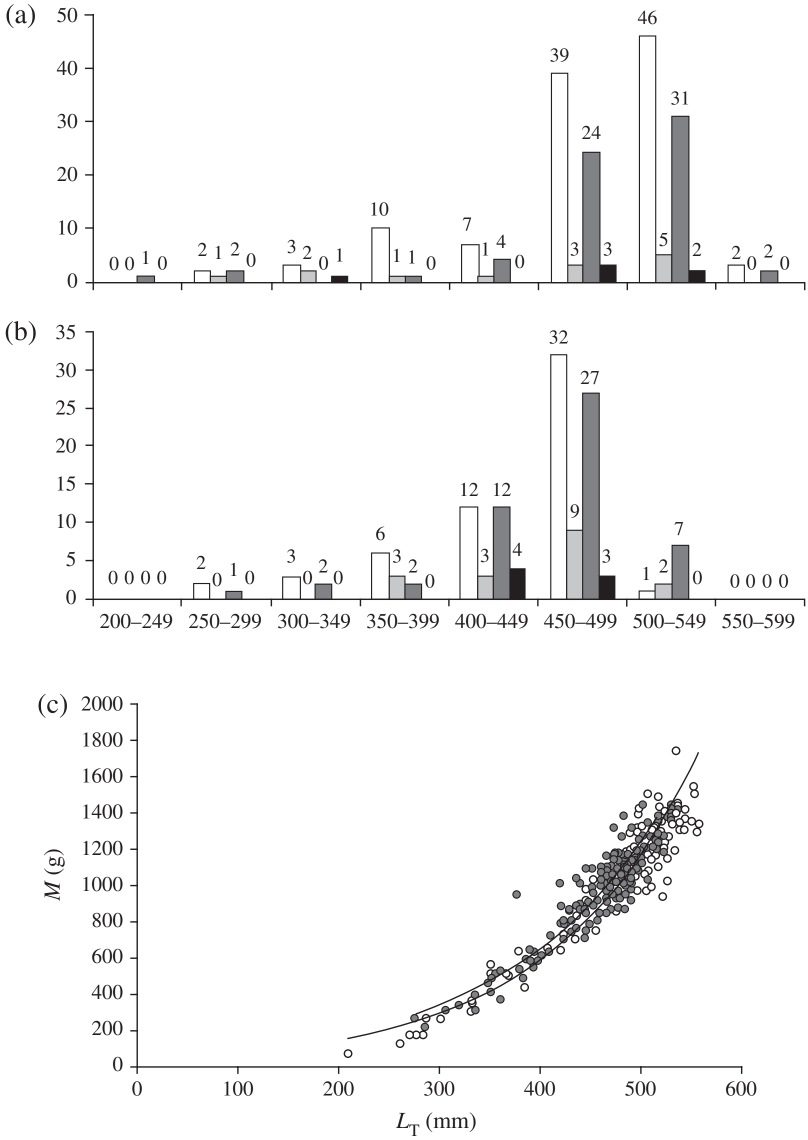

FIG. 2. Total length (L_T) per cent frequency distribution of (a) male and (b) female Amblyraja doellojuradoi by seasons; specimens caught in: □, autumn; ▒, winter; ▓, spring; ■, summer. Numbers above bars are sample sizes. (c) The relation between *A. doellojuradoi* mass (M) and L_T of both sexes: ○, males; ●, females. The curves were fitted by: female $y = 47{\cdot}3e^{0{\cdot}0066x}$ ($n = 130$, $r^2; = 0{\cdot}8646$) and male $y = 36{\cdot}2e^{0{\cdot}007x}$ ($n = 189$, $r^2; = 0{\cdot}9101$).

The logistic curve fitted for males produced an estimated $L_{T50\%}$ of 448 mm (80·5% of the observed L_{Tmax}) [Fig. 3(a)]. For females, the logistic curve fitted produced an estimated $L_{T50\%}$ of 411 mm (82·2% of the observed L_{Tmax}) [Fig. 3(b)]. No significant differences were observed in the estimated $L_{T50\%}$ between sexes ($t = 3{\cdot}76$, d.f. $= 1$, $P < 0{\cdot}05$).

TABLE I. Morphological criteria used to determine the maturity stages of *Amblyraja doellojuradoi*

Sex	Maturity stages	Macroscopic characteristics
Male	Immature (M1)	Thin and straight spermatic ducts
		Under-developed testes
		Non-calcified claspers with length shorter than pelvic fin
	Maturing (M2)	Spermatic ducts beginning to coil
		Enlarged testes starting to lobate
		Calcifying claspers as long as or a bit longer than pelvic fins
	Mature (M3)	Meandering epididymides tightly filled with sperm
		Testes with vitelline vesicles
		Calcified claspers
Female	Immature (F1)	Thin uteri
		Small ovaries with undifferentiated oocytes
		Hardly recognizable oviducal glands
	Maturing (F2)	Ovaries with transparent ovarian follicles
		Oviducal glands are visible, but are not fully developed
	Mature (F3)	Enlarged uteri
		Distended ovaries with differentiated oocytes of several sizes
		Oviducal glands fully formed
	Mature with egg capsules (F4)	Fully or partially formed egg capsule
		Large oviducal glands
		Ovaries with vitellogenic oocytes of several sizes

REPRODUCTIVE ANALYSIS

The M_T (0·94–20·00 g) increased sharply from 430 mm L_T [Fig. 4(a)], but the inflexion point of the M_T and L_T relationship was estimated at 421 mm ($P < 0{\cdot}001$, $n = 166$). The L_C in immature specimens ranged from 11 to 83 mm (mean ± S.D. = 38·9 ± 20·5 mm; $n = 27$). Individuals in maturation had L_C ranging from 9·5 to 15·4 mm (mean ± S.D. = 124·18 ± 26 mm; $n = 11$) and in mature males these values varied between 85 and 150 mm (mean ± S.D. = 126·91 ± 13·18 mm; $n = 155$). The inflexion point of the L_C and L_T relationship was estimated at 408 mm ($P < 0{\cdot}001$, $n = 193$) [Fig. 4(b)]. The N_{AT} rows varied from 0 to 4. Immature and maturing males had N_{AT} 0 to 3 and mature males mostly had N_{AT} 2 to 4 rows [Fig. 4(c)]. Specimens > 460 mm L_T and with three rows of alar thorns were all mature.

Immature females had a mean ± S.D. M_O of 3·46 ± 1·44 g (range = 1·55–5·60 g; $n = 20$), maturing females had a mean ± S.D. M_O of 8·70 ± S.D. 6·70 g (range = 3·44–21·15 g; $n = 9$) and mature females, bearing only yellow vitellogenic follicles had a mean ± S.D. M_O of 24·10 ± 9·36 g (range = 6·85–49·3 g; $n = 57$). Vitellogenic follicles in egg-bearing females occurred with corresponding mean ± S.D. M_O of 18·55 ± 8·67 g (range = 7·95–26·09 g; $n = 4$). The inflexion point of the M_O and L_T relationship was estimated at 427 mm ($P < 0{\cdot}001$, $n = 90$) [Fig. 5(a)]. The W_{OD} ranged from 2 to 50 mm and the inflexion point was estimated at 383 mm ($P < 0{\cdot}001$, $n = 122$) [Fig. 5(b)]. The W_U in mature females without capsules varied between 2 and 65 mm and the inflexion point was estimated at 400·1 mm ($P < 0{\cdot}001$, $n = 118$). The majority of mature females, however, had W_U of 10–30 mm; few females were observed with W_U

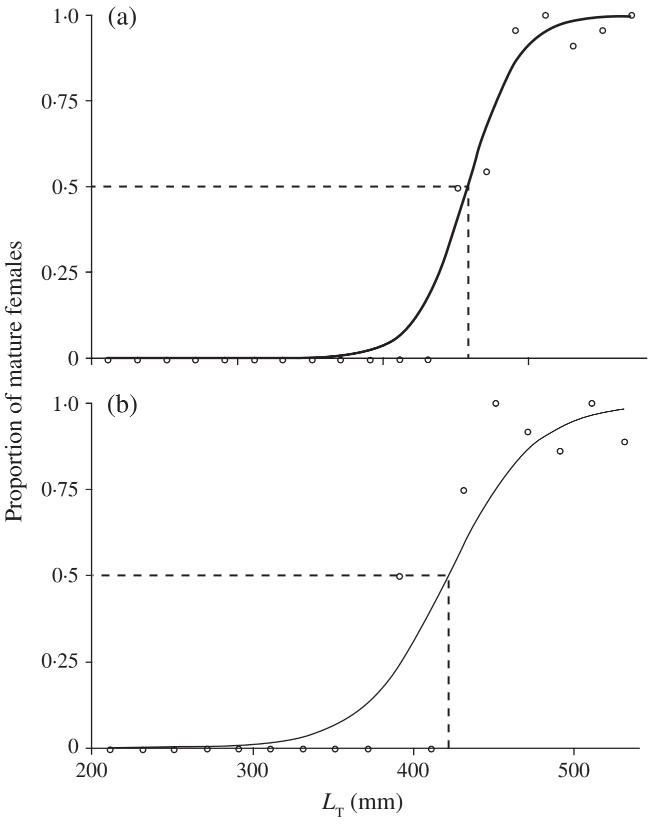

FIG. 3. Proportion of (a) immature (M1 = 0), maturing (M2 = 0·5) and mature (M3 = 1·0) male and (b) immature (F1 = 0), maturing (F2 = 0·50) and mature (F3 = 0·75 and F4 = 1·00) female *Amblyraja doellojuradoi* presented in 20 mm total length (L_T) intervals (see Table I for maturity scales). - - - -, L_T at which 50% of the specimens are sexually mature.

of 50–65 mm [Fig. 5(c)]. Seven females (432–500 mm; 750–1400 g) had capsules in their uteri, being captured during autumn ($n = 5$) and spring ($n = 2$). No significant differences between right and left M_O ($W = 2221·5$, $n = 67$, $P > 0·05$), nor in the number ($W = 1830$, $n = 63$, $P > 0·05$) or diameter ($t = -0·1052$, $n = 33$, $P > 0·05$) of ovarian follicles were found. No significant correlation between the number of ovarian follicles, which varied between 1 and 45, and the L_T of mature females (Spearman correlation = −0·1844, $n = 65$, $P > 0·05$) was found [Fig. 5(d)]. The diameter ranged from 9·2 to 35·0 mm and the correlation with L_T of mature females was not significant (Spearman correlation = 0·2155; $n = 36$; $P > 0·05$) [Fig. 5(e)]. The frequency distribution of ovarian follicles was composed of three groups: 9–18 mm (11 specimens with 112 follicles), 19–23 mm (15 specimens with 199 follicles) and 24–35 mm (10 specimens with 177 follicles) [Fig. 5(f)].

No significant differences were found in the I_H between sexes (Mann–Whitney U-test = 11614·5, d.f. = 314, $P > 0·05$). On the other hand, significant differences between maturity stages for males (Kruskal–Wallis, $\chi^2 = 12·63$, d.f. = 2, $P < 0·001$) and females were found (Kruskal–Wallis, $\chi^2 = 18·83$, d.f. = 3, $P < 0·001$). Differences

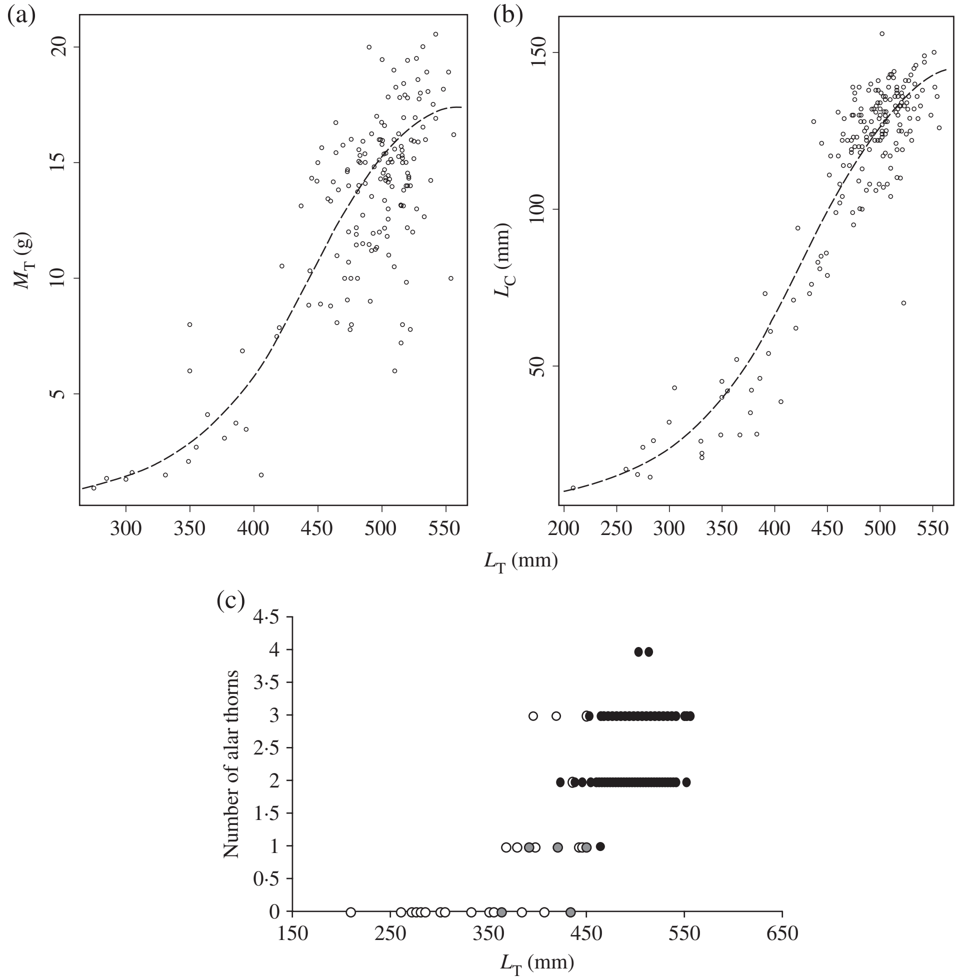

FIG. 4. Relation between total length (L_T) and (a) testes mass (M_T), (b) clasper length (L_C) and (c) number of rows of alar thorns of *Amblyraja doellojuradoi*. ○, immature; ●, maturing; ●, mature specimens.

between maturity stages in males occurred between immature (M1) and mature (Tukey test, M1 *v.* M3, $P < 0{\cdot}05$) and in females between mature, including females with ovarian capsules, and immature (Tukey test, H1 *v.* H3, $P < 0{\cdot}001$; H1 *v.* H4, $P < 0{\cdot}05$).

The I_G differed significantly between sexes (Mann–Whitney U-test = 10339·5, d.f. = 319, $P < 0{\cdot}001$) and also between sexual maturity stages for males (Kruskal–Wallis, $\chi^2 = 21{\cdot}385$, d.f. = 2, $P < 0{\cdot}001$) and females (Kruskal–Wallis, $\chi^2 = 39{\cdot}69$, d.f. = 3, $P < 0{\cdot}001$). It differed significantly between immature males and the rest of the maturity stages (Tukey test, M1 *v.* M2, $P < 0{\cdot}05$, M1 *v.* M3, $P < 0{\cdot}001$) and among immature and other female stages (Tukey test, H1 *v.* H2, $P < 0{\cdot}05$; H1 *v.* H3, $P < 0{\cdot}001$; H1 *v.* H4, $P < 0{\cdot}05$; H2 *v.* H3, $P < 0{\cdot}001$).

A significant and negative correlation between the diameter of the ovarian follicles and the I_H (Spearman correlation = −0·347, $n = 33$, $P < 0{\cdot}05$) [Fig. 6(a)] and

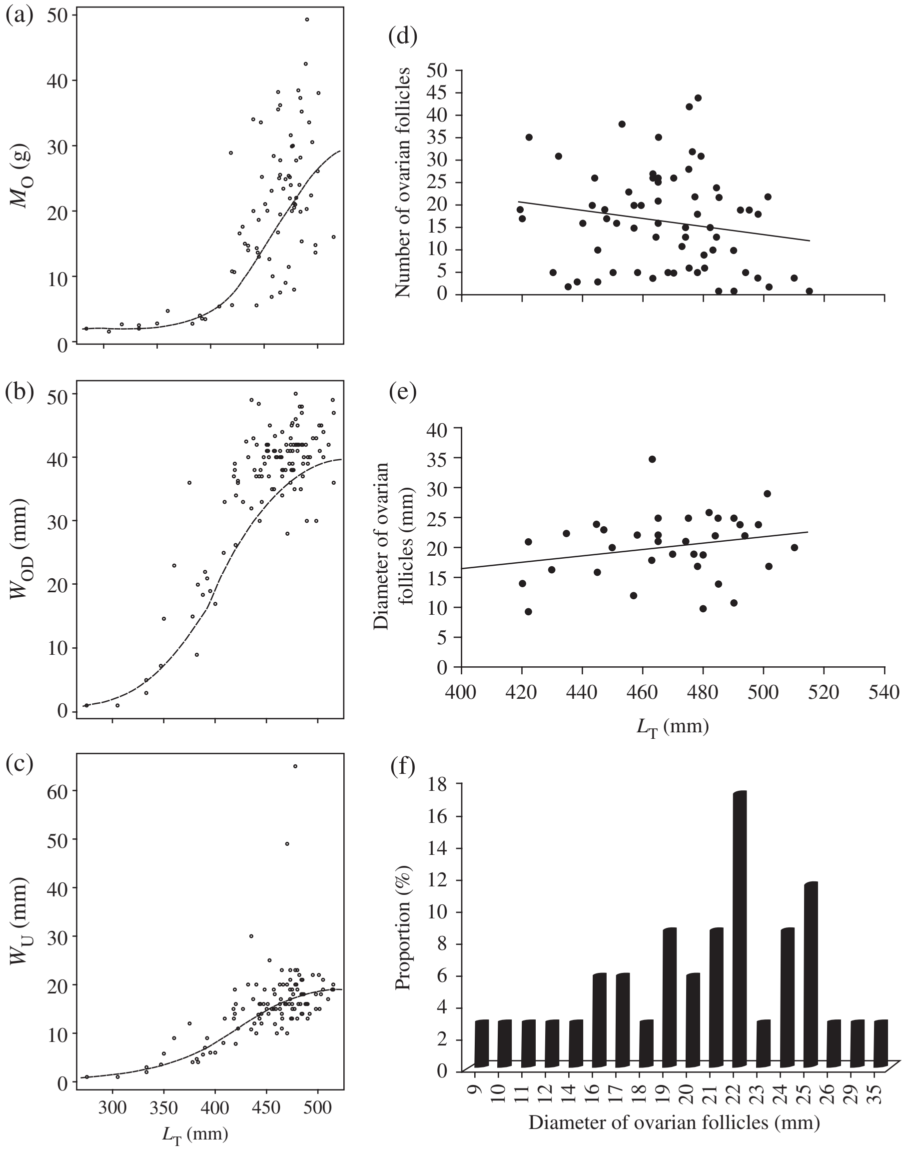

Fig. 5. Relation between total length (L_T) and (a) ovary mass (M_O), (b) oviducal gland width (W_{OD}), (c) uterus width (W_U), (d) number and (e) diameter of ovarian follicles of *Amblyraja doellojuradoi*. (f) Frequency distribution of the diameter of ovarian follicles.

a non-significant correlation among the former and the I_G (Spearman correlation = 0·0939, $n = 24$, $P > 0·05$) was found [Fig. 6(b)]. There was a negative and not significant correlation between the I_G and I_H for males (Spearman correlation = −0·1275, $P > 0·05$, $n = 190$) [Fig. 6(c)].

The average I_H between seasons was not significantly different for mature males (Kruskal–Wallis, $\chi^2 = 0·7856$, d.f. = 3, $P > 0·05$) [Fig. 7(a)], but was for mature females (Kruskal–Wallis, $\chi^2 = 23·953$, d.f. = 3, $P < 0·001$) reaching the lowest

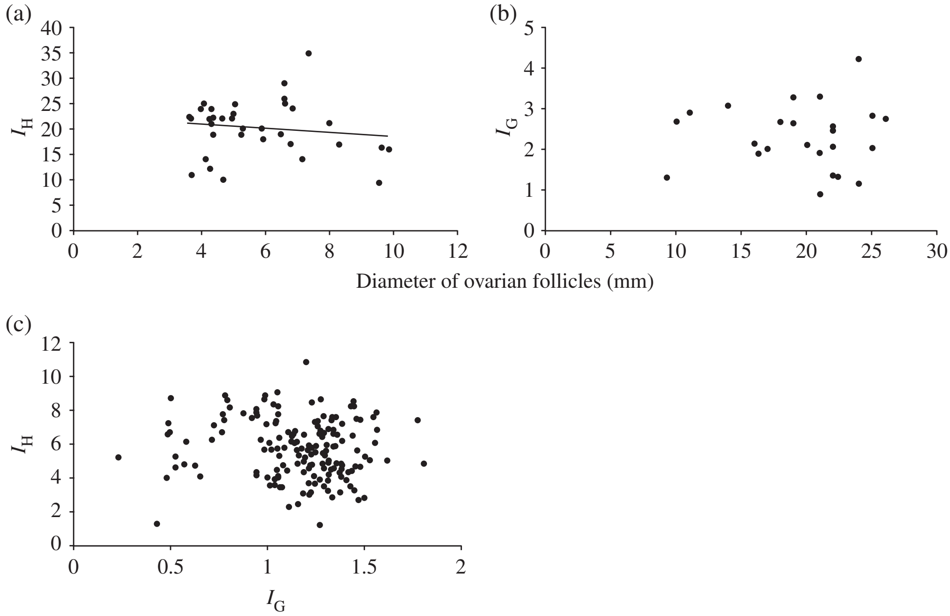

FIG. 6. Relation of the diameter of the ovarian follicles with female (a) hepato-somatic (I_H) ($y = -0{\cdot}3972x + 22{\cdot}587$) and (b) gonado-somatic (I_G) indices of *Amblyraja doellojuradoi* and (c) the relationship between I_H and I_G in males.

value in autumn (Tukey test, autumn *v*. spring, $P < 001$; autumn *v*. winter, $P < 0{\cdot}05$) [Fig. 7(b)]. The average I_G between seasons was significantly different for mature males (Kruskal–Wallis, $\chi^2 = 21{\cdot}1675$, d.f. = 3; $P < 0{\cdot}001$) [Fig. 7(c)], with the greatest value in autumn (Tukey test, autumn *v.* spring, $P < 0{\cdot}001$; autumn *v*. summer, $P < 0{\cdot}05$). In the case of mature females, I_G was nearly the same for all seasons (Kruskal–Wallis, $\chi^2 = 3{\cdot}55$, d.f. = 3, $P > 0{\cdot}05$) [Fig. 7(d)]. The diameter of the ovarian follicles did not show significant variation during the four seasons (Kruskal–Wallis, $\chi^2 = 5{\cdot}38$, d.f. = 3, $P > 0{\cdot}05$) [Fig. 7(e)]. The W_{OD} differed between seasons (Kruskal–Wallis, $\chi^2 = 14{\cdot}19$, d.f. = 3, $P < 0{\cdot}01$) having the highest values in females captured in winter (Tukey test, winter *v.* autumn, $P < 0{\cdot}05$; winter *v.* summer, $P < 0{\cdot}01$) [Fig. 7(f)]. The number of ovarian follicles also varied between seasons (Kruskal–Wallis, $\chi^2 = 22{\cdot}82$, d.f. = 3, $P < 0{\cdot}001$), with the greatest number in autumn (Tukey test, autumn *v*. winter, $P < 0{\cdot}001$; autumn *v*. spring, $P < 0{\cdot}05$; autumn *v*. summer, $P < 0{\cdot}05$) [Fig. 7(g)].

DISCUSSION

Dimensions and mass of fishes are often used to characterize the growth, sexual maturation, the maximum size and population structure (Francis, 2006). Marked increases of the parameters of some sexual characteristics match perfectly with the estimated average L_T at sexual maturity. The M_T and L_C, and the M_O and W_{OD}, are good indicators of sexual maturity. Nevertheless, some characteristics such as alar thorns in males and W_U are not useful for determining sexual maturation. These features have a wide

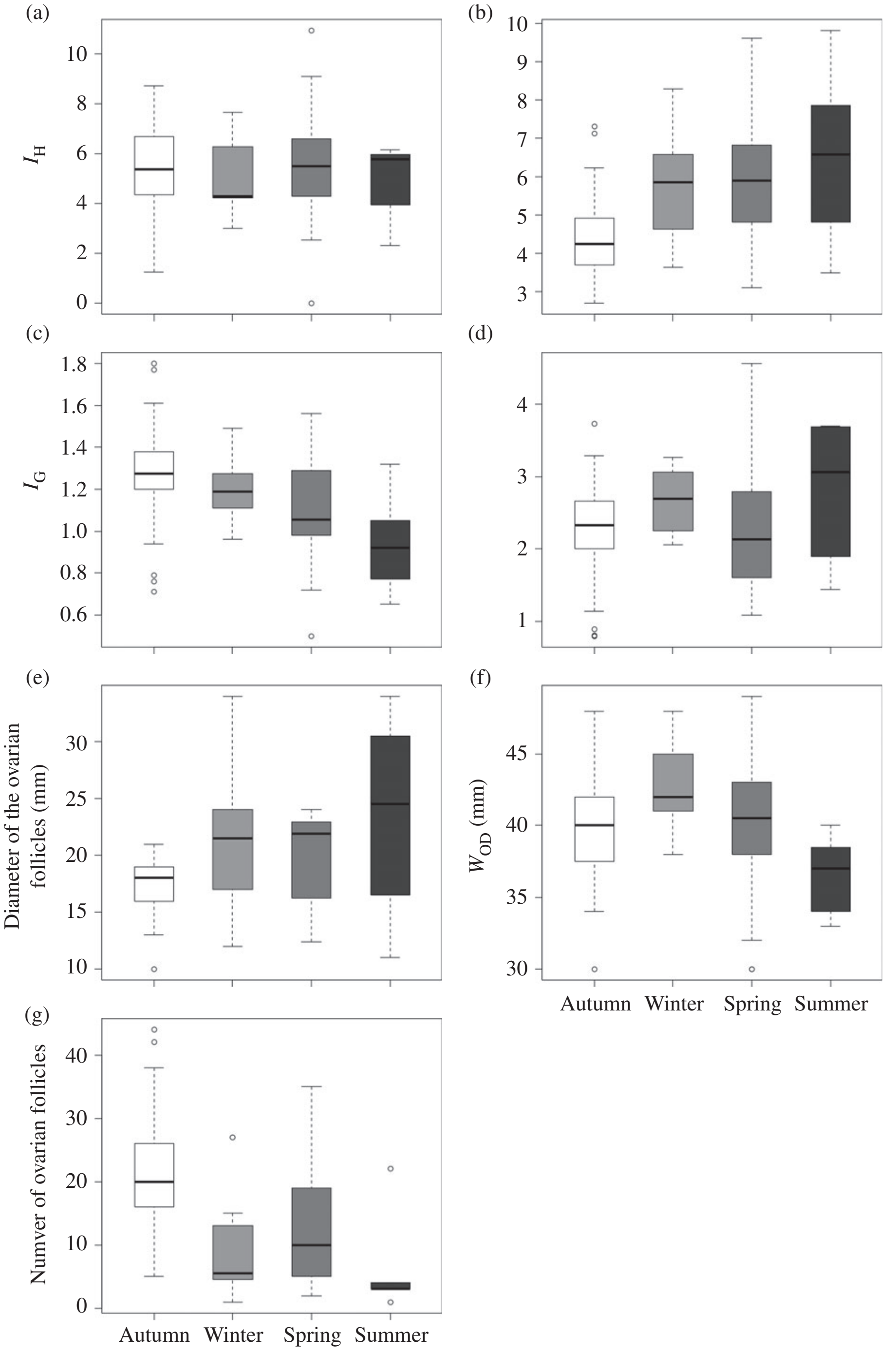

FIG. 7. Seasonal (a, b) hepato-somatic (I_H) and (c, d) gonado-somatic (I_G) indices for mature (a, c) male and (b, d) female *Amblyraja doellojuradoi*. Seasonal (e) diameter of the ovarian follicles, (f) oviducal gland width (W_{OD}) and (g) the number of ovarian follicles are also shown. The line inside the boxes indicates the mean, the boxes the S.E. and between brackets indicate the number of specimens analysed.

range of variation that is not consistent with a given L_T. Males use the alar thorns to hold the females during copulation (McEachran & Konstantinou, 1996); however, although the number of alar thorn rows increases with L_T, they can also be present in juveniles. The W_U can be misinterpreted in specimens that recently released egg capsules, making this measurement inaccurate.

Generally, female skates attain larger size as a consequence of their reproductive strategy (Walmsley-Hart *et al.*, 1999). This occurs in the majority of species of the family Rajidae (Ebert, 2005; Oddone & Vooren, 2005; Oddone *et al.*, 2005, 2008 Colonello *et al.*, 2007*b*; Scenna, 2011). In the present work, however, it was males of *A. doellojuradoi* were larger than females, although $L_{T50\%}$ estimation did not significantly differ between sexes. One possible explanation for this difference between sexes could be that at a certain point females stop investing their energy in growth in favour of reproductive processes. Elasmobranchs generally mature at 75% of their maximum size (Cortés, 2000). Holden (1974) noted that most elasmobranchs mature at between 60 and 90% of L_{Tmax}, indicating an extensive juvenile stage; followed by a brief maturing stage and a short period of sexually maturity (Ebert, 2005). The mean L_T at first maturity for males and females of sympatric skate species [*Bathyraja albomaculata* (Norman 1937), *Bathyraja brachyurops* (Fowler 1910), *Bathyraja macloviana* (Norman 1937), *Bathyraja magellanica* (Philippi 1902), *Psammobatis bergi* Marini 1932, *Psammobatis extenta* (Garman 1913) and *R. agassizii*] compared to *A. doellojuradoi* was between 70 and 80% of the maximum L_T observed (Braccini & Chiaramonte, 2002; San Martín *et al.*, 2005; Ruocco *et al.*, 2006; Colonello *et al.*, 2007*b*). The same species in different parts of the world can reach $L_{T50\%}$ at different lengths (Templeman, 1987; Colonello *et al.*, 2007*b*, *c*). *Amblyraja radiata* (Donovan 1808) has different maturity estimations in different areas (Templeman, 1987; Sulikowski *et al.*, 2005*a*). Templeman (1987) reported a bimodal pattern of maturity in both sexes for *A. radiata* on the Scotland Eastern Platform, with some mature individuals substantially smaller than other sizes. Given the wide depth inhabited by *A. radiata* [18–966 m; McEachran & Musick (1975)], differences in $L_{T50\%}$ could be reflecting different patterns of species habitat use (Sulikowski *et al.*, 2005*a*). In this regard, specimens of *A. doellojuradoi* were captured from 75 to 414 m depth, half the depth range recorded for *A. radiata*. Therefore, it is likely that the habitat is not sufficiently heterogeneous to show a change in size at maturity in *A. doellojuradoi*.

In elasmobranchs it is usual to find sexual dimorphism in liver size, it being generally larger in females (Rossouw, 1987). The sex differences may be related to increased energy demand representing ovarian follicle maturation (Lucifora, 2003), since the liver plays an important role in the female reproductive cycle, storing lipids and participating in vitellogenesis by producing vitellogenin, the precursor of the yolk (Koob & Callard, 1999). In this regard, *A. doellojuradoi* has no correlation between the I_H and the diameter of ovarian follicles, but it is important to note that it would be necessary to collect more data to confirm the correlation analysis of the ovarian follicles diameter in relation to different variables. An I_H difference between the sexes is reported for other species inhabiting the Argentinean continental shelf such as *S. bonapartii* (Mabragaña *et al.*, 2002), *R. agassizii* (Estalles *et al.*, 2009), *Atlantoraja cyclophora* (Regan 1903) (M. C. Oddone, unpubl. data) and *Atlantoraja platana* (Günther 1880) (A. S. Marcal, unpubl. data). Mature females of these skate species have a higher value than males or immature females. In *A. doellojuradoi* I_H differences were observed during ontogeny in both males and females, but no differences between sexes of mature individuals were found. This similarity in the I_H between sexes is unusual in skates, but has been described in *Raja clavata* L. 1758 (Saglam & Ak, 2012) and *A. radiata* (Sulikowski *et al.*, 2005*a*). Moreover, liver size may vary during ontogeny of males and females (Mabragaña *et al.*, 2002). The greater mass of the liver of adults compared with immature males is related to the metabolic needs of the formation of gametes during the breeding season, migration to areas of mating and starvation resistance (Rossouw, 1987). From the analysis of sex ratio, the hypothesis of migration to mating areas is strengthened, since a higher proportion of mature males was found in autumn. It is already known that unequal sex ratios in chondrichthyan populations may be a consequence of sexual segregation (Springer, 1967). Analysing the distribution of the specimens by seasons, however, it was observed that there is not spatial segregation, so the idea of possible migration to a reproductive area must be discarded. Therefore, supplementary information is required to determine the reason that males are more frequent in the cold season.

Regarding the variation of I_H through the year, no significant differences were observed between seasons in males, but in females the lowest I_H was found in autumn. The lack of monthly samples makes it difficult to determine whether the variation of I_H in females is due to the seasonal cycles of reproduction or may be caused by deposit or differential lipid storage. In males, the stable value of I_H and no correlation with the I_G indicate that the liver is probably continuously storing and metabolizing lipids without causing significant changes in its biomass, exerting a metabolic function and indicating possible reproduction throughout the year (Sulikowski *et al.*, 2005*a*, *b*).

The I_G is a good indicator of elasmobranch reproduction, since mating is correlated with the I_G (Yamaguchi *et al.*, 1997, 2000; Kyne & Bennett, 2002; Yamaguchi & Kume, 2009). This index was significantly higher in females than males of *A. doellojuradoi*. According to Capapé *et al.* (2007), this feature is related to the continuous production of vitellogenic oocytes and is indicative of continuous reproductive activity throughout the year. In females of *A. doellojuradoi* no significant differences between the I_G among seasons were observed, suggesting that they would be prepared to breed throughout the year. This observation is supported by the lack of seasonal variation in the diameter of ovarian follicles. Males showed seasonal variations with the highest I_G in autumn. Other species also show seasonal differences in the values of the I_G, *e.g. P. bergi* (San Martín *et al.*, 2005), *R. clavata* (Saglam & Ak, 2012) and *Leucoraja naevus* (Müller & Henle 1841) (Du Buit, 1976).

Formation of egg capsules may be seasonal or occur throughout the year (Hamlett & Koob, 1999). In coastal waters, temperature and light can directly influence reproductive activity (Holden, 1975), generating reproductive seasonal peaks. This has been suggested for *S. bonapartii* (Mabragaña *et al.*, 2002), *S. acuta* (Mabragaña *et al.*, 2014), *P. bergi* (San Martín *et al.*, 2005), *P. extenta* (Braccini & Chiaramonte, 2002) and *R. agassizii* (Colonello, 2009). Other rajids, such as *Bathyraja* spp. (Ruocco *et al.*, 2006; Scenna, 2011) inhabiting deep waters, lack these peaks in reproductive activity. The deep-water skate *A. radiata* has a single annual cycle (Sulikowski *et al.*, 2005*a*). The reproductive cycle of *A. doellojuradoi* is difficult to estimate since no information on monthly reproductive activity is available. The information provided by the ovarian follicles, the I_G and I_H, however, can be used to determine the periodicity of egg-capsule production since they are good indicators of the regularity of the reproductive cycle (Jons & Miranda, 1997; Yamaguchi *et al.*, 1997, 2000; Kyne & Bennett, 2002; Yamaguchi & Kume, 2009). Gathering all the information, it could be concluded that females had the lowest I_H in autumn probably due to investment of lipids in reproduction. The higher I_G in males in the same season would indicate that have increased sperm production. The growth of the W_{OD} in winter and the rise in the number of ovarian follicles in the same period reaffirm the existence of a strong reproductive peak in autumn. Therefore, it might be concluded that *A. doellojuradoi* is able to reproduce throughout the year, with the peak reproductive period in autumn when females were captured with egg capsules. This is consistent with the hypothesis of continuous reproductive cycles at depth.

Knowledge of the reproductive biology, the frequency of reproduction, growth parameters and also the estimated fishing mortality are critical for the development of management plans for skate populations, since skates are highly susceptible to fishing pressure (Walker, 1998; Walker & Hislop, 1998; Dulvy & Reynolds, 2002). This group is caught as by-catch in trawl fisheries and can be discarded at sea or landed as by-catch (Massa & Hozbor, 2003; Cedrola *et al.*, 2005; Tamini *et al.*, 2006). Survival from discarding is only known for the area of the Falkland (Malvinas) Islands (Laptikhovsky, 2004), but not from the Argentinean continental shelf. Chondrichthyan landings (especially *Mustelus schmitti* Springer 1939 and skates) have steadily increased recently (Massa *et al.*, 2004; Cousseau *et al.*, 2007) and currently the fisheries data show a general decrease in relative biomass of skates on the Argentinean continental shelf (Massa & Hozbor, 2003). To achieve sustainable management of *A. doellojuradoi* and other skate species it is necessary to know the details of landings, fishing effort statistics, a better understanding of the population structure, vital population parameters and possible latitudinal clines. Principal aspects of the life history of *A. doellojuradoi* are still unknown and more complementary studies are necessary to determine adequate management plans for the species.

I would like to thank the Instituto Nacional de Investigación y Desarrollo Pesquero (INIDEP) and R. Gonzalez for collecting specimens from different research and commercial cruises, the Ciencias Marinas department of the Universidad Nacional de Mar del Plata for the workplace and finally C. Spath, M. D. Antoni, J. M. Díaz de Astarloa and L. Lucifora for the critical reading that improved the quality of this work. G.D. was supported by a scholarship from the Consejo Nacional de Investigaciones Científicas y Tecnológicas (CONICET).

References

Aubone, A. & Wohler, O. C. (2000). Aplicación del método de máxima verosimilitud a la estimación de parámetros y comparación de curvas de crecimiento de Von Bertalanffy. *INIDEP Informe Técnico Interno N°* **37**.

Braccini, J. M. & Chiaramonte, G. E. (2002). Reproductive biology of *Psammobatis extenta*. *Journal of Fish Biology* **61,** 272–288.

Camhi, M., Fowler, S., Musick, J., Bräutigam, A. & Fordham, S. (1998). Sharks and their relatives: ecology and conservation. *Occasional Paper of the IUCN Species Survival Commission* **20,** 1–39.

Capapé, C., Guelorget, O., Siau, Y., Vergne, Y. & Quignard, J. P. (2007). Reproductive biology of the thornback ray *Raja clavata* (Chondrichthyes: Rajidae) from the coast of Languedoc (Southern France, Northern Mediterranean). *Vie et Milieu* **57,** 83–90.

Cedrola, P. M., González, A. M. & Pettovello, A. D. (2005). Bycatch of skates (Elasmobranchii: Arhynchobatidae, Rajidae) in the Patagonian red shrimp fishery. *Fisheries Research* **71,** 141–150.

Colonello, J. H. (2009). Ecología reproductiva de tres batoideos (Chondrichthyes): *Atlantoraja castelnaui* (Rajidae), *Rioraja agassizii* (Rajidae) y *Zapteryx brevirostris* (Rhinobatidae).

Implicancias de distintas estrategias adaptativas en un escenario de explotación intensiva. PhD Thesis, Universidad Nacional de La Plata, Argentina.

Colonello, J. H., Christiansen, H. E. & Macchi, G. J. (2007a). Escala de madurez sexual para peces cartilaginosos de la Plataforma Continental Argentina. *INIDEP Informe Técnico Interno N°* **74**.

Colonello, J. H., García, M. L. & Lasta, C. A. (2007b). Reproductive biology of *Rioraja agassizii* from the coastal southwestern Atlantic ecosystem between northern Uruguay (34°) and northern Argentina (42° S). *Environmental Biology of Fishes* **80,** 277–284.

Colonello, J. H., Lucifora, L. O. & Massa, A. M. (2007c). Reproduction of the angular shark (*Squatina guggenheim*): geographic differences, reproductive cycle, and sexual dimorphism. *ICES Journal of Marine Science* **64,** 131–140.

Cortés, E. (2000). Life history patterns and correlations in sharks. *Reviews in Fisheries Science* **8,** 299–344.

Cousseau, M. B., Figueroa, D. E., Díaz de Astarloa, J. M., Mabragaña, E. & Lucifora, L. O. (2007). *Rayas, chuchos y otros batoideos del Atlántico Sudoccidental (34° S–55° S)*. Mar del Plata:INIDEP.

Crawley, M. J. (2005). *Statistics: An Introduction Using R*. Chichester: John Wiley & Sons.

Du Buit, M. H. (1976). The ovarian cycle of the cuckoo ray, *Raja naevus* (Müller & Henle), in the Celtic Sea. *Journal of Fish Biology* **8,** 199–207.

Dulvy, N. K. & Reynolds, J. D. (2002). Predicting extinction vulnerability in skates. *Conservation Biology* **16,** 440–450.

Dulvy, N. K., Metcalfe, J. D., Glanville, J., Pawson, M. G. & Reynolds, J. D. (2000). Fishery stability, local extinctions, and shifts in community structure in skates. *Conservation Biology* **14,** 283–293.

Ebert, D. A. (2005). Reproductive biology of skates, *Bathyraja* (Ishiyama), along the eastern Bering Sea continental slope. *Journal of Fish Biology* **66,** 618–649.

Enever, R., Catchpole, T. L., Ellis, J. R. & Grant, A. (2009). The survival of skates (Rajidae) caught by demersal trawlers fishing in UK waters. *Fisheries Research* **97,** 72–76.

Estalles, M., Perez Comezaña, J. E., Tamini, L. L. & Chiaramonte, G. E. (2009). Reproductive biology of the skate, *Rioraja agassizii* (Müller & Henle, 1841), off Puerto Quequén, Argentina. *Journal of Applied Ichthyology* **25,** 60–65.

Francis, M. P. (2006). Morphometric minefields – towards a measurement standard for chondrichthyan fishes. *Environmental Biology of Fishes* **77,** 407–421.

García, V. B., Lucifora, L. O. & Myers, R. A. (2008). The importance of habitat and life history to extinction risk in sharks, skates, rays and chimaeras. *Proceedings of the Royal Society B* **275,** 83–89. doi: 10.1098/rspb.2007.1295

Hamlett, W. C. & Koob, T. J. (1999). Female reproductive systems. In *Sharks, Skates and Rays: The Biology of Elasmobranch Fishes* (Hamlett, W. C., ed), pp. 398–443. Baltimore, MD: Johns Hopkins University Press.

Holden, M. J. (1974). Problems in the rational exploitation of elasmobranchs populations and some suggested solutions. In *Sea Fisheries Research* (Harden Jones, F. R., ed), pp. 117–137. London: Paul Elek.

Holden, M. J. (1975). The fecundity of *Raja clavata* in British waters. *Journal du Conseil pour l'Exploration de la Mer* **36,** 110–118.

Jons, G. D. & Miranda, L. E. (1997). Ovarian weight as an index of fecundity, maturity, and spawning periodicity. *Journal of Fish Biology* **50,** 150–156.

Koob, T. J. & Callard, I. A. (1999). Reproductive endocrinology of female elasmobranchs: lessons from the little skate (*Raja erinacea*) and spiny dogfish (*Squalus acanthias*). *Journal of Experimental Zoology* **284,** 557–574.

Kyne, P. M. & Bennett, M. B. (2002). Diet of the eastern shovelnose ray, *Aptychotrema rostrata* (Shaw & Nodder, 1794), from Moreton Bay, Queensland, Australia. *Marine and Freshwater Research* **53,** 679–686.

Laptikhovsky, V. V. (2004). Survival rates for rays discarded by the bottom trawl squid fishery off the Falkland Islands. *Fisheries Bulletin* **102,** 757–759.

Lasta, C. A., Ruarte, C. O. & Carozza, C. R. (2001). Flota costera argentina: antecedentes y situación actual. In *El Mar Argentino y sus Recursos Pesqueros. Tomo 3: Evolución de la Flota Pesquera Argentina, Artes de Pesca y Dispositivos Selectivos* (Bertolotti, M. I., Verazay, G. A. & Akselman, R., eds), pp. 89–119. INIDEP : Mar del Plata.

Lucifora, L. O. (2003). Ecología y conservación de los grandes tiburones costeros de Bahía Anegada, Provincia de Buenos Aires. PhD Thesis, Universidad Nacional de Mar del Plata, Argentina.

Mabragaña, E. & Cousseau, M. B. (2004). Reproductive biology of two sympatric skates in the Southwest Atlantic: *Psammobatis rudis* (Günther, 1870) and *P. normani* (McEachran, 1983). *Journal of Fish Biology* **65,** 559–573.

Mabragaña, E., Lucifora, L. O. & Massa, A. M. (2002). The reproductive ecology and abundance of *Sympterygia bonapartii* endemic to the south–west Atlantic. *Journal of Fish Biology* **60,** 951–967.

Mabragaña, E., Lucifora, L. O., Corbo, M. de. L. & Díaz de Astarloa, J. M. (2014). Seasonal reproductive biology of the bignose fanskate *Sympterygia acuta* (Chondrichthyes, Rajidae). *Estuaries and Coasts* **38,** 1466–1476.

Massa, A. M. & Hozbor, N. M. (2003). Peces cartilaginosos de la Plataforma Argentina, explotación, situación y necesidades para un manejo adecuado. *Publicación de la Comisión Técnica Mixta del Frente Marítimo* **19,** 199–206.

Massa, A. M., Lucifora, L. O. & Hozbor, N. M. (2004). Condrictios de la región costera bonaerense y uruguaya. In *El Mar Argentino y sus Recursos Pesqueros. Los Peces Marinos de Interés Pesquero Caracterización Biológica y Evaluación del Estado de Explotación* (Boschi, E. E., ed), pp. 85–99. Mar del Plata : INIDEP.

Matta, M. E. & Gunderson, D. R. (2007). Age, growth, maturity, and mortality of the Alaska skate, *Bathyraja parmifera*, in the eastern Bering Sea. *Environmental Biology of Fishes* **80,** 309–323. doi: 10.1007/s10641-007-9223-8

McEachran, J. D. & Konstantinou, H. (1996). Survey of the variation in alar and malar thorns in skates: phylogenetic implications (Chondrichthyes: Rajoidei). *Journal of Morphology* **228,** 165–178.

McEachran, J. D. & Musick, J. A. (1975). Distribution and relative abundance of seven species of skates (Pisces: Rajidae) which occur between Nova Scotia and Cape Hatteras. *Fishery Bulletin* **73,** 110–136.

Mcphie, R. P. & Campana, S. E. (2009). Reproductive characteristics and population decline of four species of skate (Rajidae) off the eastern coast of Canada. *Journal of Fish Biology* **75,** 223–246. doi: 10.1111/j.1095-8649.2009.02282.x

Musick, J. A. & Ellis, J. K. (2005). Reproductive evolution of chondrichthyans. In *Reproductive Biology and Phylogeny of Chondrichthyes: Sharks, Batoids and Chimaeras* (Hamlett, W. C., ed), pp. 45–80. Enfield, NH: Enfield Science Publishers Incorporated.

Oddone, M. C. & Vooren, C. M. (2005). Reproductive biology of *Atlantoraja cyclophora* (Regan 1903) (Elasmobranchii: Rajidae) off southern Brazil. *ICES Journal of Marine Science* **62,** 1095–1103.

Oddone, M. C., Paesch, A. F. & Norbis, W. (2005). Size at first sexual maturity of two species of rajoi skates, genera *Atlantoraja* and *Dipturus* (Pisces, Elasmobranchii: Rajidae), from the south–western Atlantic Ocean. *Journal of Applied Ichthyology* **21,** 70–72.

Oddone, M. C., Amorim, A. F., Mancini, P. L., Norbis, W. & Velasco, G. (2007). The reproductive biology and cycle of *Rioraja agassizii* (Müller & Henle, 1841) (Chondrichthyes, Rajidae), in southeast Brazil, SW Atlantic Ocean. *Scientia Marina* **71,** 593–604.

Oddone, M. C., Velasco, G. & Rincon, G. (2008). Occurrence of freshwater stingrays (Chondrichthyes: Potamotrygonidae) in the Uruguay River and its tributaries, Uruguay, South America. *Aqua, International Journal of Ichthyology* **14,** 69–76.

Roa, R., Ernst, B. & Tapia, F. (1999). Estimation of size at sexual maturity: an evaluation of analytical and resampling procedures. *Fishery Bulletin US* **97,** 570–580.

Rossouw, G. J. (1987). Function of the liver and hepatic lipids of the lesser sand shark, *Rhinobatos annulatus* (Müller & Henle). *Comparative Biochemistry and Physiology B* **86,** 785–790.

Ruocco, N. L., Lucifora, L. O., Díaz de Astarloa, J. M. & Wöhler, O. (2006). Reproductive biology and abundance of the white–dotted skate, *Bathyraja albomaculata*, in the southwest Atlantic. *ICES Journal of Marine Science* **63,** 105–116.

Saglam, H. & Ak, O. (2012). Reproductive biology of *Raja clavata* (Elasmobranchii: Rajidae) from southern Black Sea coast around Turkey. *Helgoland Marine Research* **66,** 117–126.

San Martín, M. J., Perez Comezaña, J. E. & Chiaramonte, G. E. (2005). Reproductive biology of the south west Atlantic marble sand skate *Psammobatis bergi* Marini, 1932 (Elasmobranchii, Rajidae). *Journal of Applied Ichthyology* **21,** 504–510.

Scenna, L. B. (2011). Biología y ecología reproductiva de las especies del género *Bathyraja* (Elasmobranchii: Rajidae) en la Plataforma Continental Argentina. PhD Thesis, Universidad Nacional de Mar del Plata, Argentina.

Scenna, L. B. & Díaz de Astarloa, J. M. (2014). Reproductive biology of the Magellan skate, *Bathyraja magellanica* (Chondrichthyes, Rajidae), in the south-western Atlantic. *Marine and Freshwater Research* **65,** 766–775. doi: 10.1071/MF13144

Springer, S. (1967). Social organization of shark populations. In *Sharks, Skates and Rays* (Gilbert, P. W., Mathewson, R. F. & Rall, D. P., eds), pp. 149–174. Baltimore, MD: Johns Hopkins University Press.

Stevens, J. D., Bonfil, R., Dulvy, N. K. & Walker, P. A. (2000). The effects of fishing on sharks, rays, and chimaeras (Chondrichthyans), and the implications for marine ecosystem. *ICES Journal of Marine Science* **57,** 476–494.

Sulikowski, J. A., Kneebone, J., Elzey, S., Jurek, J., Danley, P. D., Howel, W. H. & Tsang, P. W. C. (2005a). The reproductive cycle of the thorny skate (*Amblyraja radiata*) in the western Gulf of Maine. *Fishery Bulletin* **103,** 536–543.

Sulikowski, J. A., Tsang, P. W. C. & Howel, W. H. (2005b). Age and size at sexual maturity for the winter skate, *Leucoraja ocellata*, in the western Gulf of Maine based on morphological, histological and steroid hormone analyses. *Environmental Biology of Fishes* **72,** 429–441.

Tamini, L. L., Chiaramonte, G. E., Perez, J. E. & Cappozzo, H. L. (2006). Batoids in a coastal fishery of Argentina. *Fisheries Research* **77,** 326–332.

Templeman, W. (1987). Differences in sexual maturity and related characteristics between populations of thorny skate (*Raja radiata*) in the Northwest Atlantic. *Journal of Northwest Atlantic Fisheries Science* **7,** 155–167.

Walker, T. I. (1998). Can shark resources be harvested sustainably? A question revisited with a review of shark fisheries. *Marine and Freshwater Research* **49,** 553–572.

Walker, P. A. & Hislop, J. R. G. (1998). Sensitive skates or resilient rays? Spatial and temporal shifts in ray species composition in the central and north–western North Sea between 1930 and the present day. *ICES Journal of Marine Science* **55,** 392–402.

Walmsley-Hart, S. A., Sauer, W. H. H. & Buxton, C. D. (1999). The biology of skates *Raja wallacei* and *R. pullopunctata* (Batoidea: Rajidae) on the Agulhas Bank, South Africa. *South African Journal of Marine Science* **21,** 165–179.

Winemiller, K. O. & Rose, K. A. (1992). Patterns of life–history diversification in North American fishes: implications for population regulation. *Canadian Journal of Fisheries and Aquatic Sciences* **49,** 2196–2218.

Yamaguchi, A. & Kume, G. (2009). Reproductive biology of the fanray, *Platyrhina sinensis* (Batoidea: Platyrhinidae) in Ariake Bay, Japan. *Ichthyological Research* **56,** 133–139. doi: 10.1007/s10228-008-0078-6

Yamaguchi, A., Taniuchi, T. & Shimizu, M. (1997). Reproductive biology of the starspotted dogfish *Mustelus manazo* from Tokyo Bay, Japan. *Fisheries Science* **63,** 918–922. doi: 10.2331/fishsci.63.918

Yamaguchi, A., Taniuchi, T. & Shimizu, M. (2000). Geographic variations in reproductive parameters of the starspotted dogfish, *Mustelus manazo*, from five localities in Japan and in Taiwan. *Environmental Biology of Fishes* **57,** 221–233. doi: 10.1023/A:1007558324902

Zar, J. H. (1984). *Biostatistical Analysis*. New Jersey, NJ: Prentice–Hall Inc.