

Trace elements in the statoliths of jumbo flying squid off the Exclusive Economic Zones of Chile and Peru

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ABSTRACT: Ontogenetic variation in 4 trace element (⁸⁸Sr, ¹³⁷Ba, ²⁴Mg, ²³Na) concentrations and their ratios to Ca were measured in statoliths of the jumbo flying squid *Dosidicus gigas* off the Exclusive Economic Zone of Chilean and Peruvian waters using laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS). The element compositions of statoliths showed no significant differences between females and males. All of the elements in different growth zones showed significant variations, except for Mg. Sr:Ca and Mg:Ca were good indicators for distinguishing squid from autumn and winter spawning seasons. Sr:Ca and Ba:Ca distribution patterns in statoliths confirmed that paralarvae and juvenile squid inhabit surface waters, while subadult squid migrate into deeper waters. An increasing Sr:Ca ratio of subadult squid could be explained by declining temperature gradients from northern to southern sampling locations, although no significant Sr:Ca differences were observed ($p > 0.05$). Mg:Ca ratios decreased progressively from the nucleus to the peripheral zone, which might be correlated with statolith growth rates. Na:Ca ratios slightly declined from paralarvae to the subadult phase. Quantitative relationships between statolith trace elements and environmental conditions under different growth stages are needed to improve our understanding of life history of *D. gigas*.

KEY WORDS: *Dosidicus gigas* · Life history · Chile · Peru · Strontium · Barium · Magnesium · Sodium

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INTRODUCTION

The jumbo flying squid *Dosidicus gigas* (d'Orbigny, 1835) is the most abundant and the largest omastrephid widely distributed in the eastern Pacific Ocean from California (37° N) to southern Chile (47° S); its distribution extends westward to 125° W around the Equator (Nesis 1983, Nigmatullin et al. 2001). The highest abundance is found between the Gulf of California and waters off northern Chile, especially off the Peruvian coast (Nigmatullin et al. 2001). *D. gigas* supports one of the most important cephalopod fisheries in

the eastern Pacific Ocean. During 2004 to 2008, annual global catches of *D. gigas* were about 787 000 t, mainly by Peru, Mexico, Chile and China. The Chinese squid jigging fleet began targeting *D. gigas* in 2002, with annual landings reaching 50 000 to 205 000 t during 2002 to 2008.

In Chilean waters, *Dosidicus gigas* are landed year-round, supporting 1 of the 3 cephalopod fisheries (i.e. *Loligo gahi*, *Octopus mimus*, and *D. gigas*; Rocha 1997, Rocha & Vega 2003, Zúñiga et al. 2008). *D. gigas* are mainly concentrated in waters off central Chile, where the estimated biomass in 2005 and 2006 was over

130 000 t (Alarcón-Muñoz et al. 2008). Spatial and temporal changes in size structure and reproductive activity of *D. gigas* were analyzed in the central-southern area off Chile (34° to 40° S), and variability may have resulted from migration of the species (Ibáñez & Cubillos 2007). In waters outside the Exclusive Economic Zone (EEZ), *D. gigas* have been targeted by the Chinese squid jigging vessels since 2004 (Chen & Zhao 2005). Three scientific surveys for the squid were made during 2006 to 2008 (Liu et al. 2010b). In Peruvian waters, many studies have been conducted to improve our understanding of the basic fisheries biology of *D. gigas* (e.g. Argüelles et al. 2001, Tafur et al. 2001, Taípe et al. 2001).

Cephalopod statoliths are calcified structures within the paired statocysts, which provide a sense of balance (Arkhipkin & Bizikov 2000) and also act as an acceleration receptor; the statocyst provides much information about biology and ecology over an organism's life span (Arkhipkin 2005). Therefore, statoliths are perhaps best suitable for analyzing age and growth of cephalopods (Jackson 1994). Recently, geochemical signatures based on composition of trace elements and isotopes of calcified structures have become increasingly popular as a tool for investigating population structure and habitats (Thorrold et al. 2002), and are widely used in coral skeletons, bivalve shells, gastropods, and fish otoliths (Beck et al. 1992, Campana 1999, Zacherl et al. 2003a, Becker et al. 2005). Statoliths share many similarities in geochemistry and structure with fish otoliths, and the elemental composition of statoliths may represent a useful source of information on cephalopod habitats (Clarke 1978, Radtke 1983, Arkhipkin et al. 2004).

The chemical composition of cephalopod statoliths is important for discriminating different populations, analyzing life history, and reconstructing habitat environmental conditions (Liu et al. 2010a). Statolith signatures have been used to distinguish stocks of the Patagonian longfin squid *Loligo gahi* (Arkhipkin et al. 2004) and to determine life histories of seven-star flying squid *Martialia hyadesi* (Rodhouse et al. 1994), Japanese common squid *Todarodes pacificus* (Ikeda et al. 1996, 2003), neon flying squid *Ommastrephes bartrami* (Yatsu et al. 1998), veined squid *L. forberi* (Biemann & Piatkowski 2001), and boreoatlantic armhook squid *Gonatus fabricii* (Zumholz 2005). Previous results have shown the association of different elements with environmental conditions, especially water temperature; some of these elements (e.g. Sr, Fe, and Zn) are considered 'thermometers' for reconstructing environment temperature. For *T. pacificus*, a negative relationship is observed between Sr concentration in statoliths and water temperature, whereas Fe and Zn show positive relation-

ships (Ikeda et al. 1996). Several trace element concentrations in *Dosidicus gigas* statoliths were measured by particle induced X-ray emission (PIXE) in Peruvian and Costa Rican waters during El Niño and non-El Niño years, with no significant difference being found in Sr concentration despite higher water temperatures in El Niño years (Ikeda et al. 2002b). However, the difference in the Sr concentration between samples from the 2 waters is positively related to salinity (Ikeda et al. 2002b).

In our study we applied laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) to determine element concentrations and ratios of calcium, strontium, barium, magnesium and sodium in the different zones of *Dosidicus gigas* statoliths collected from the waters outside EEZs of Chile and Peru, to quantify the composition and spatial distribution of these elements in the statolith, and to compare these elements among squid spawning in different seasons and collected from different locations. The information derived can improve our understanding of the life history of *D. gigas*.

MATERIALS AND METHODS

Squid collection. Fourteen specimens of *Dosidicus gigas* were sampled by the Chinese jigging fleet during scientific surveys from 2007 to 2008 in waters outside the EEZ of Chile, and another 4 were collected by industrial vessels in the waters outside the EEZ of Peru (Fig. 1, Table 1). Squid were measured on board for mantle length (ML) to the nearest 1 mm and body weight (BW) to the nearest 1 g. Sex was identified and maturity stages were determined by eye on a scale of 1 to 5 (Lipinski 1979). Statoliths were dissected and stored in 90% alcohol for further analysis. A detailed description of all specimens used in this study is given in Table 1.

Age estimation and hatching date calculation. In total, 18 paired statoliths were extracted. Only the left one, if not damaged, was prepared for age estimation and element analysis. We followed standard aging methodology (Dawe & Natsukari 1991). The number of increments for each statolith was counted independently by 2 readers. The average was used if the range of counts was within 10% of the mean (Yatsu et al. 1997).

Element analysis. After aging, the statolith sections were rinsed in MilliQ water for further element analysis. Several spots of analysis were set from the core to the edge of the dorsal dome for each statolith (Fig. 2). The first point was placed in the nucleus (N), while the second point was in the postnuclear zones (PN), representing the embryonic and larval phases,

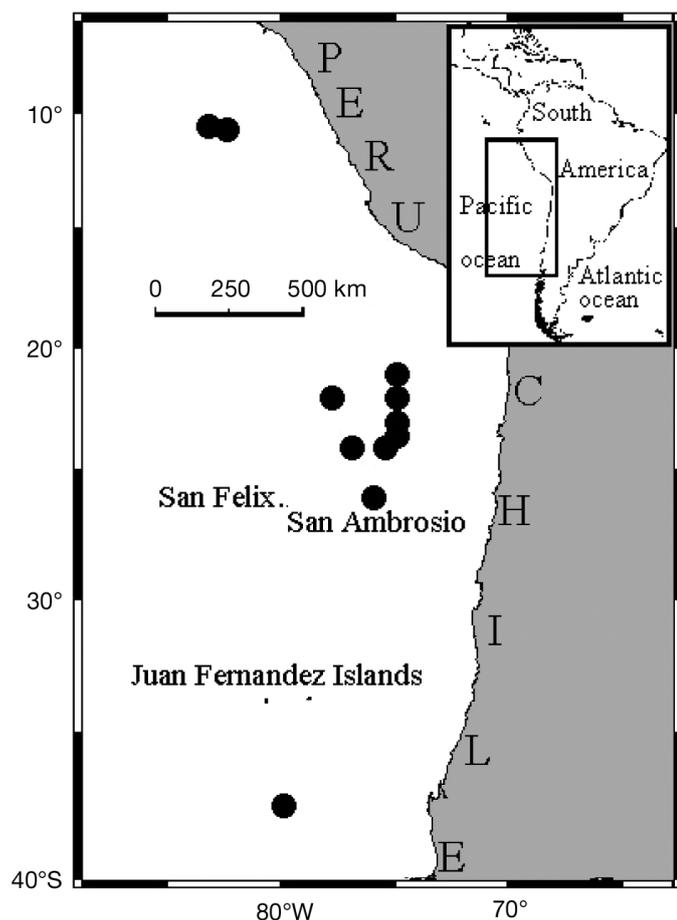


Fig. 1. *Dosidicus gigas*. Sampling locations (●) off the Exclusive Economic Zones of Chile and Peru

respectively (Arkhipkin 2005). Two to 4 points were in the dark zone (DZ), representing the juvenile phase, and 1 to 5 points were placed in the peripheral zone (PZ), representing the subadult phase (Arkhipkin 2005).

Five elements (^{43}Ca , ^{88}Sr , ^{137}Ba , ^{24}Mg , ^{23}Na) were determined by LA-ICP-MS at the State Key Laboratory of Geological Processes and Mineral Resources, China University of Geosciences, using a 193 nm frequency-quadrupled ArF excimer laser coupled with Agilent 7500a. Ablation parameters in the analyses included a spot size of 24 μm and a laser pulse of 5 Hz. Helium (0.7 l min^{-1}) was used as the sample gas in the ablation cell, and Argon (0.8 l min^{-1}) was subsequently added to the gas flow. Every spot in the statoliths was ablated for 20 s to determine 5 elements.

Statistical analysis. Two-way analysis of variance (ANOVA) was used to test for significant differences in element:Ca ratios among specimens and between the statolith growth zones.

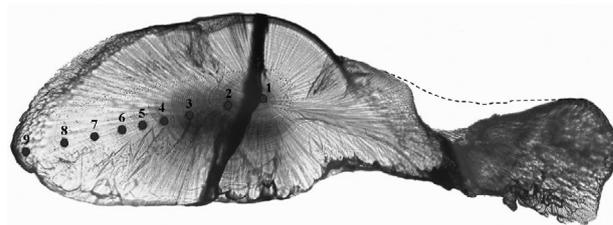


Fig. 2. *Dosidicus gigas*. Ground statolith, showing spots of analysis: 1 = nucleus; 2 = postnuclear zone; 3 to 6 = dark zone; and 7 to 9 = peripheral zone

Table 1. *Dosidicus gigas*. Summary information of sampled squid. SST: sea surface temperature, ML: mantle length, BW: body weight, nd: no data, ♀: female, ♂: male. Maturity was based on a scale of 1 to 5 following Lipinski (1979). Dates are given as yyyy/mm/dd

No.	Sample date	Sample area	Sample location	SST (°C)	ML (mm)	BW (g)	Sex	Maturity stage	Age (d)	Spawning date	Spawning season
6b	2007/01/20	Central	77°00'W, 24°00'S	20.84	391	1800	♀	1	237	2006/05/28	Autumn
23b	2007/01/21	Central	76°00'W, 26°00'S	21.44	425	2250	♀	1	251	2006/05/15	Winter
35b	2007/01/25	South	80°00'W, 37°30'S	18.21	368	1350	♀	1	201	2006/07/08	Winter
38b	2007/01/25	South	80°00'W, 37°30'S	18.21	309	800	♀	1	207	2006/07/02	Autumn
61a	2008/05/01	Central	75°00'W, 23°00'S	nd	381	1643	♀	1	216	2007/09/28	Spring
69b	2008/05/01	Central	75°00'W, 23°00'S	nd	361	1302	♂	1	235	2007/09/09	Spring
88a	2008/05/07	Central	77°50'W, 22°00'S	nd	449	2735	♂	2	242	2007/09/08	Spring
75b	2008/05/07	Central	77°50'W, 22°00'S	nd	517	3719	♀	2	191	2007/10/29	Spring
321a	2008/05/09	Central	75°30'W, 24°00'S	nd	372	1535	♀	1	217	2007/10/05	Spring
316a	2008/05/11	Central	75°00'W, 23°30'S	nd	341	1338	♂	1	248	2007/09/06	Spring
294a	2008/05/12	Central	75°00'W, 22°00'S	nd	391	1978	♀	1	184	2007/11/10	Spring
301a	2008/05/12	Central	75°00'W, 22°00'S	nd	401	2127	♀	1	291	2007/07/26	Winter
164b	2008/05/12	Central	75°00'W, 22°00'S	nd	402	1789	♀	1	202	2007/10/23	Spring
49b	2008/05/14	Central	75°00'W, 21°00'S	nd	373	1457	♂	1	209	2007/10/18	Spring
124b	2008/09/13	North	82°29'W, 10°39'S	nd	122	533	♀	1	159	2007/04/07	Autumn
140b	2008/09/13	North	82°29'W, 10°39'S	nd	275	549	♀	1	164	2007/04/02	Autumn
61b	2008/09/20	North	83°14'W, 10°32'S	nd	258	456	♀	1	159	2007/04/07	Autumn
70b	2008/09/20	North	83°14'W, 10°32'S	nd	291	735	♀	1	176	2007/03/28	Autumn

Table 2. *Dosidicus gigas*. Element concentrations and element:Ca ratios in statoliths analyzed using laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS), means \pm SD

Element	Concentration (ppm)		Element:Ca ratio	
	Range	Mean	Range	Mean
Strontium	5871–6570	6186 \pm 213	14.8–16.4 mmol mol ⁻¹	15.6 \pm 0.5
Barium	4.5–9.5	6.7 \pm 1.5	11.2–23.8 μ mol mol ⁻¹	16.8 \pm 3.9
Magnesium	30.9–93.9	61.3 \pm 19.3	79–233 μ mol mol ⁻¹	154 \pm 48
Sodium	3382–5115	4174 \pm 584	8.6–12.8 mmol mol ⁻¹	10.5 \pm 1.4

Table 3. *Dosidicus gigas*. Analysis of variance of the element concentrations in different growth zones of statoliths. N: nucleus, PN: postnuclear zone, DZ: dark zone, PZ: peripheral zone

Element	Range of element concentrations (ppm)				p
	N	PN	DZ	PZ	
Strontium	5477–7218	5487–6868	5572–6275	5968–7433	0.038
Barium	3.5–10.5	3.7–13.9	3.8–9.4	5.1–15.7	0.004
Magnesium	32.3–383	29.1–94.2	26.6–68.7	22.7–547	0.427
Sodium	3589–5239	3616–5519	3380–5388	2982–4421	0.000

RESULTS

Age estimation and hatching date calculation

All 18 individuals were immature, ranging from 258 to 517 mm ML and 456 to 3719 g BW (Table 1). Based on statolith aging, the smallest squid was 159 d old, while the oldest was 291 d. Back-calculated hatching dates of the samples included autumn (March, April, May), spring (September, October, November), and winter cohorts (July; Table 1).

Element concentrations and element:Ca ratios

In addition to Ca, Sr was the second-most abundant element, with concentrations ranging from 5871 to 6570 ppm in the statoliths. The ratio Sr:Ca ranged from 14.8 to 16.4 mmol mol⁻¹. Other element:Ca ratios are shown in Table 2. The element composition of statoliths showed no significant differences between females and males ($p > 0.05$). All elements in different growth zones showed significant variations, except for Mg (Table 3).

Element:Ca ratios in different growth zones and age classes

Sr:Ca ratios were higher in the N, then decreased gradually to become lowest in the DZ and increased towards the PZ (Fig. 3a). Values in the N and PZ were significantly higher than in the DZ ($p < 0.05$).

Ba:Ca ratios in the PZ were significantly higher than those in the other zones (Figs. 3b & 4b; $p < 0.01$). No significant difference was found between the N and DZ ($p > 0.05$).

Mg:Ca ratios showed a decreasing trend along the transect from the core to the edge (Fig. 3c), and their values in the N were significantly higher than those in the other zones ($p < 0.01$). Na:Ca ratios in the PZ were significantly lower than those in the other zones (Fig. 3d; $p < 0.01$). No significant differences in Na:Ca ratios were found between the other zones ($p > 0.05$).

The changes in element:Ca ratios with age for individual squid (Fig. 4) were almost the same as those in different growth zones.

Element:Ca ratios in different hatching seasons

The Sr:Ca ratio in the N was the only ratio found to be significantly different for squid hatching in the winter and autumn (Fig. 5a; $p < 0.05$). There were no significant differences of other element:Ca ratios in *Dosidicus gigas* statoliths from the 3 spawning seasons ($p > 0.05$). No significant difference of Sr:Ca ratios in the PN was found among the 3 spawning seasons ($p > 0.05$), although their seasonal patterns were similar to those in the N (Fig. 5a). Ba:Ca ratios in the PN showed no significant difference among seasons (Fig. 5b; $p > 0.05$).

Mg:Ca ratios in the N of the autumn hatched squid were significantly higher than those of squid hatched in other seasons (Fig. 5c; $p < 0.05$). The values of Mg:Ca in the other growth zones showed no significant differences between squid hatched in different seasons (Fig. 5c; $p > 0.05$).

The 4 growth zones in statoliths of squid hatched in different seasons showed no significant effects on Na:Ca (Fig. 5d; $p > 0.05$).

Element:Ca ratios in PZ from different sampling locations

Sr:Ca ratios in statoliths in the subadult phase increased with decreased latitude, but no significant differences were found between them (Fig. 6a, $p > 0.05$). Ba:Ca and Mg:Ca had a similar distribution in statoliths from northern to southern locations, with values from the central waters being higher than those from northern and southern waters (Fig. 6b,c). Na:Ca

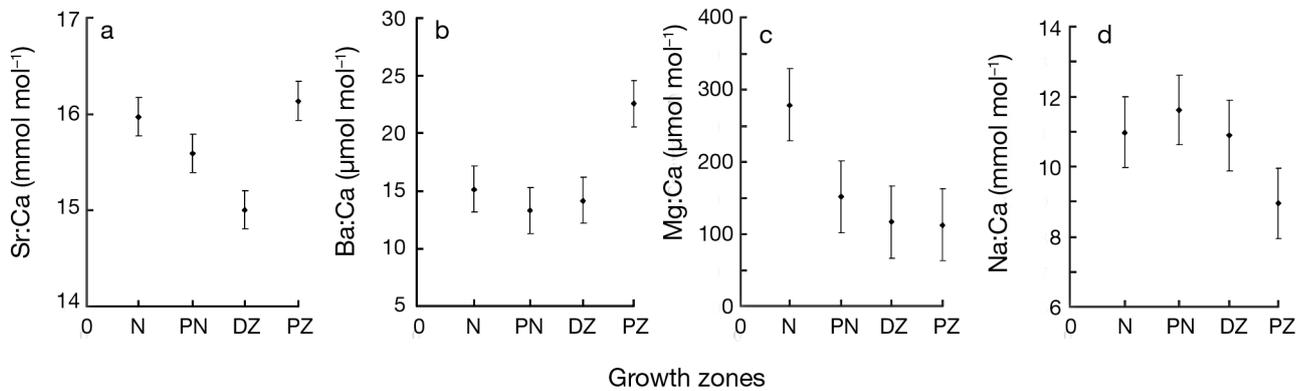


Fig. 3. *Dosidicus gigas*. Element:Ca ratios measured with laser ablation inductively coupled plasma mass spectrometry (LA-ICP-MS) in different growth zones of statoliths from the nucleus to the edge. N: nucleus, PN: postnuclear zone, DZ: dark zone, PZ: peripheral zone

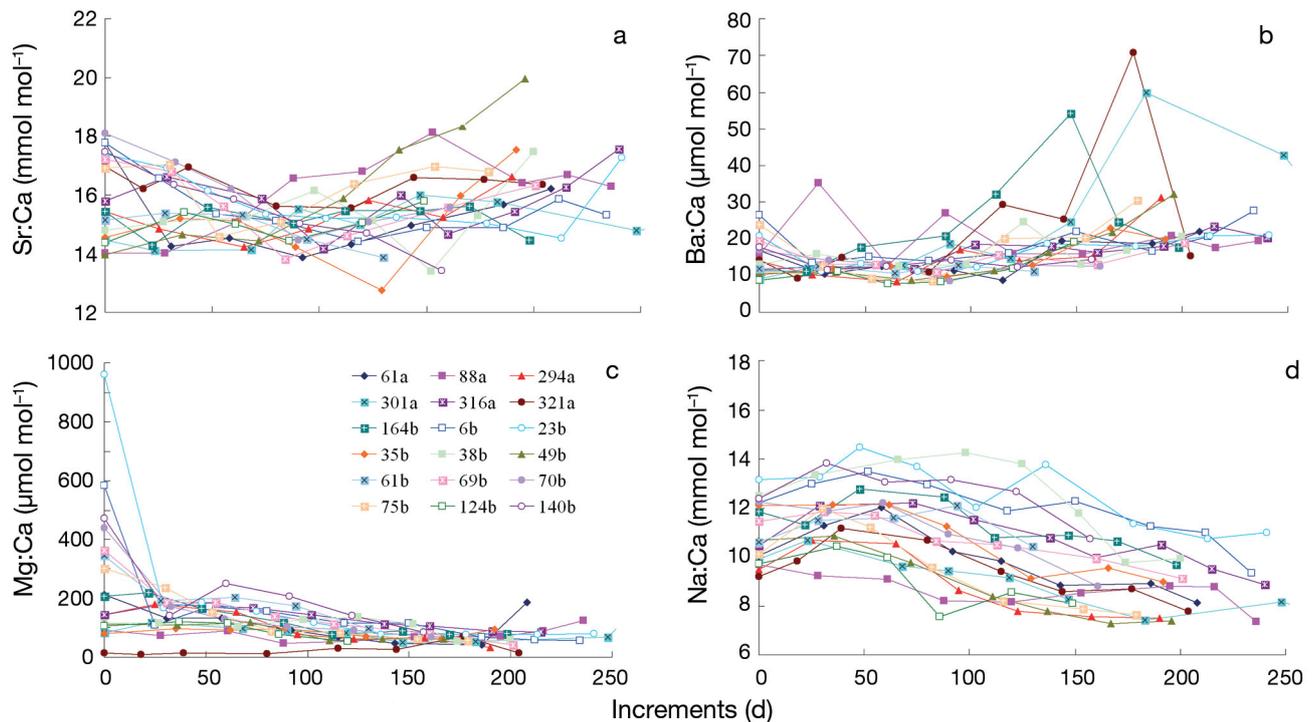


Fig. 4. *Dosidicus gigas*. Changes in element:Ca ratios measured with LA-ICP-MS with age (days). Each symbol represents 1 squid (see key in panel (c) and Table 1)

values in statoliths collected from northern, central, and southern waters were almost the same (Fig. 6d, $p > 0.05$).

DISCUSSION

Statolith microstructure of ommastrephid squids comprises the nucleus (N), postnuclear zone (PN), dark zone (DZ), and peripheral zone (PZ; Lipinski

1991). The formation of these zones generally corresponds with major ontogenetic stages. The N is the area within hatching increments that are associated with the embryonic stage (Arkhipkin 2005). The PN is associated with the larval phase of ontogenesis, the DZ represents the juvenile phase and the PZ corresponds to the subadult and adult phases (Arkhipkin & Perez 1998, Arkhipkin 2005). Evidence supporting that the PN represents the larval stage of *Dosidicus gigas* was provided by Arkhipkin & Mur-

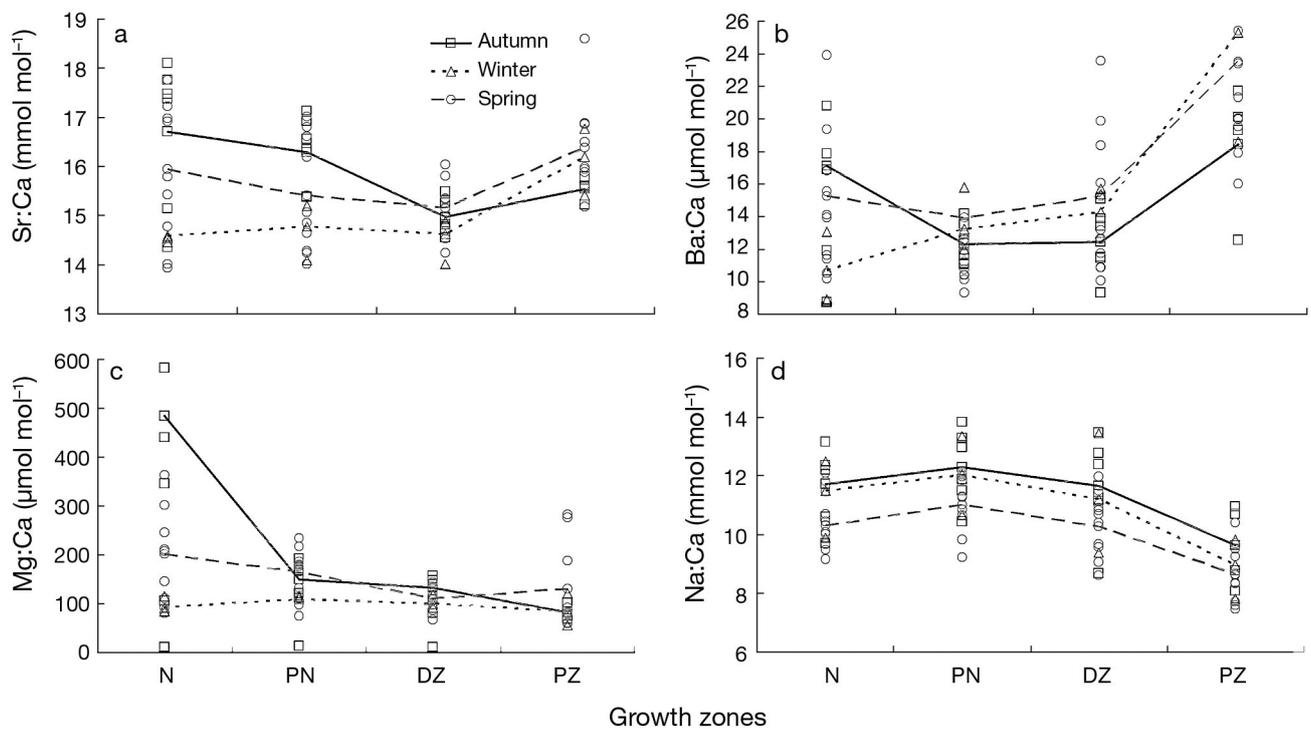


Fig. 5. *Dosidicus gigas*. Element:Ca ratios measured with LA-ICP-MS in different growth zones during different hatching seasons. For abbreviations, see Fig. 3

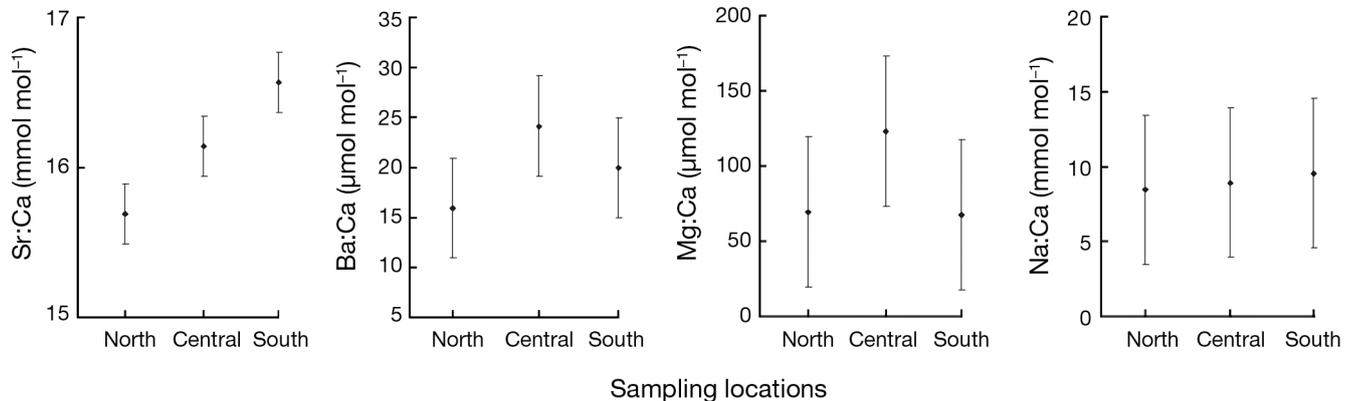


Fig. 6. *Dosidicus gigas*. Element:Ca ratios measured with LA-ICP-MS in the peripheral zone of statoliths from squid sampled in different locations

zov (1986). According to these previous studies, we analyzed the variability in element:Ca ratios among different ontogenetic stages of *D. gigas* to reconstruct the environmental history these sampled squid had experienced.

In past years, microbeam methods of PIXE and the electron probe micro-analyzer (EPMA) have generally been used to analyze classified structure micro-chemistry (Ikeda et al. 1996, 2002a). However, these studies focussed on Sr, the most abundant element incorporated into the aragonite matrix of statoliths. Recently, more scientists have paid attention to other

trace elements like Ba, Mg, and Mn, which cannot be detected accurately by these 2 methods. In this study, we used LA-ICP-MS to examine the elemental signature of *Dosidicus gigas* statoliths, since this method has a higher spatial resolution and lower detection limits.

Sr is a key element responsible for the deposition of cephalopod statoliths (Lipinski 1993). Sr:Ca ratios in statoliths of *Dosidicus gigas* (14.8 to $16.4 \text{ mmol mol}^{-1}$) were higher than those of other squid such as *Todarodes pacificus* (8.5 to 10 mmol mol^{-1} ; Ikeda et al. 2003), *Loligo gahi* ($\sim 8.0 \text{ mmol mol}^{-1}$; Arkhipkin et al. 2004), and *Gonatus fabricii* (6.3 to $8.1 \text{ mmol mol}^{-1}$; Zumholz

2005). We observed a U-shaped pattern of Sr:Ca, similar to *Ommastrephes bartrami* (Yatsu et al. 1998) and *G. fabricii* (Zumholz et al. 2007b).

Sr:Ca ratios are believed to have negative correlations with ambient temperature (Arkhipkin et al. 2004). In our study, Sr:Ca ratios in statoliths of the embryonic stage were higher than those of paralarvae and juveniles, but same as those for the subadult stage (Fig. 3a). Sr:Ca ratios of paralarvae and juvenile periods were relatively lower compared to those of the subadult stage, which is consistent with the life history of *Dosidicus gigas*. *D. gigas* are commonly found in from the surface to at least 1200 m depth (Nigmatullin et al. 2001), and the paralarvae and juvenile squid are found near the sea surface with relatively higher average ambient temperatures (upper temperature limits range from 15 to 32°C in surface waters (Nigmatullin et al. 2001). During the subadult and adult period, they migrate into the mesopelagic layers with lower temperatures (not lower than 4.0 to 4.5°C; Nigmatullin et al. 2001), although they also occur in the superficial waters at night for feeding.

We assumed that the specimens had already inhabited the feeding ground and did not migrate into the spawning ground before maturity since they were all subadults (>150–180 mm ML, except 1 specimen of 122 mm ML; Yatsu et al. 1999) and immature (maturity stage 1 or 2). This suggests that no massive horizontal migrations occurred prior to the capture of the specimens used in this study. Excluding environmental changes brought about by horizontal migration, Sr:Ca ratios in the statoliths of subadults from different sampling locations, i.e. northern part from 10° to 12° S, central part from 21° to 26° S, and southern part (south to 37° S), were determined. An increasing Sr:Ca ratio may be explained by a decrease in temperature from northern to southern waters in the study area, although no significant Sr:Ca differences were observed in the squid from different locations ($p > 0.05$). This result is similar to those of Ikeda et al. (2002b), who reported that Sr concentrations in *Dosidicus gigas* statoliths showed no significant difference between El Niño and non-El Niño years in Peruvian and Costa Rican waters for deep diel vertical migrations leading to widely ambient temperature variations.

Small variations may be worth considering, despite no significant Sr:Ca differences in larvae, juvenile, and subadult statoliths of squid hatching in the 3 seasons ($p > 0.05$). Among squid spawning in the 3 seasons, the Sr:Ca ratio of larval squid spawning in autumn was highest, whereas the value was lowest in the subadult stages. Thus, we conclude that different stages might experience different ambient water environments.

The embryonic stage occurs before hatching, and individuals at this stage depend on their yolks. Thus, at

this stage, the element signature of the statolith N is perhaps more likely to depend on squid from the same cohort, less affected by external factors because yolk composition may be influenced by genetic factors (Zumholz 2005). Yatsu et al. (1998) revealed that Sr and Ca concentrations in statoliths of *Ommastrephes bartrami* are probably affected in a different way during the embryonic period than during other ontogenetic stages. Bustamante et al. (2002) pointed out that the capsule membrane of *Sepia officinalis* acts as an efficient shield, protecting the embryo against metals like Zn and Cd. The higher Sr:Ca ratio during the embryonic period may not be due to ambient water conditions but rather to the yolk sac, as has been reported in salmonids (Kalish 1990) and *Gonatus fabricii* (Zumholz et al. 2007b). We found that Sr:Ca ratios in statoliths during the embryonic stage were different among squid spawning in the 3 seasons, but significant variation was only observed between autumn- and winter-spawning groups (Fig. 4a; $p < 0.05$). Thus, we can use Sr:Ca to discriminate the squid hatching seasons.

Ba:Ca ratios in statoliths of *Dosidicus gigas* (11.2 to 23.8 $\mu\text{mol mol}^{-1}$) were higher than those reported for *Gonatus fabricii* (5.7 to 8.2 $\mu\text{mol mol}^{-1}$; Zumholz 2005) and *Loligo gahi* (3.0 to 8.0 $\mu\text{mol mol}^{-1}$; Arkhipkin et al. 2004). This variability may result from species differences rather than from other factors. Ba concentrations are considered to increase with depth. For corals and cephalopods, Ba has therefore been used as an indicator element for upwelling events (Lea et al. 1989, Arkhipkin et al. 2004). Arkhipkin et al. (2004) suggested that intensification of upwelling is responsible for the elevated levels of Ba in *L. gahi* statoliths collected from shelf waters around the Falkland Islands. Positive correlations of Ba:Ca in gastropod larval protoconchs (Zacherl et al. 2003b) and fish otoliths (Bath et al. 2000) with Ba:Ca in the seawater were identified. Alternatively, Ba:Ca was considered a useful indicator for vertical movements of cephalopods (Arkhipkin et al. 2004). Thus an elevating trend of Ba:Ca in *D. gigas* statoliths can be confirmed by ontogenetic vertical migrations (OVMs) from the surface during paralarval and juvenile phases to mesopelagic waters in the subadult and adult periods. Similar results were found in *G. fabricii* statoliths (Zumholz 2005). Ba:Ca cannot be used to discriminate the 3 different spawning groups because there is no significant difference in different ontogenetic phases of *D. gigas*. However, it is useful to determine the autumn and spring spawning groups of *L. gahi* (Arkhipkin et al. 2004). Ba:Ca ratios in *L. gahi* (Arkhipkin et al. 2004) and *Sepia officinalis* (Zumholz et al. 2007a) statoliths were found to be negatively related to water temperature, but this was not found for *D. gigas* in our study.

Mg is another important element for biomineralization in cephalopod statoliths, and its high concentrations are considered to be associated with the deposition of organic components into the statoliths (Morris 1991). It therefore would be expected to decrease correspondingly to the size of the cephalopod (Bettencourt & Guerra 2000). Zumholz (2005) suggested that the Mg:Ca ratios of *Gonatus fabricii* were high in core regions and declined progressively towards the edge, probably reflecting decreasing growth rates from the core to the edge of the statoliths. In this study, similar to the Mg:Ca distribution for *G. fabricii*, the values of Mg:Ca gradually decreased from the N to the PZ. Arkhipkin et al. (2004) suggested that similar to coral skeletons, Mg:Ca in *Loligo gahi* statoliths was positively correlated with ambient water temperatures (Mitsuguchi et al. 1996). However, no such a relationship was found for *Dosidicus gigas* in our study. Like Sr:Ca, Mg:Ca can also be used to help identify whether squid hatched in autumn or winter.

Contrary to other element:Ca ratios, Na:Ca ratios in statoliths of *Dosidicus gigas* (8.6 to 12.8 mmol mol⁻¹) were lower compared to values for *Gonatus fabricii* (15 to 22 μmol mol⁻¹; Zumholz 2005). Na:Ca ratios declined slightly from the paralarvae to adult phases. However, no significant variations were observed in squid hatched in different seasons, suggesting that the Na:Ca ratio cannot be used to distinguish individuals hatched in different seasons.

In conclusion, Sr:Ca and Mg:Ca of *Dosidicus gigas* statoliths were good indicators for distinguishing squid from autumn and winter spawning seasons. Distribution patterns of Sr:Ca and Ba:Ca in statoliths confirm that paralarvae and juvenile squid inhabit surface waters, while subadult squid migrate into deeper waters. An increasing Sr:Ca ratio of subadult squid could be explained by declining temperature gradients from northern to southern sampling locations. Mg:Ca ratios decreased progressively from the nucleus to the peripheral zone, which may be correlated with statolith growth rates. Elemental signatures can improve our understanding of the life history of *D. gigas*.

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