Life history traits of the giant squid *Architeuthis dux* revealed from stable isotope signatures recorded in its beaks

Ángel Guerra[1], Alejandro Rodríguez-Navarro[2,] Ángel F. González[1], C. S. Romanek[3], Pedro Álvarez-Lloret[2] and Graham. J. Pierce[4]

1. Departamento de Recursos y Ecología Marina. Instituto de Investigaciones Marinas (CSIC), Vigo, Spain.
2. Departamento de Mineralogía y Petrología, Universidad de Granada, Granada, Spain.
3. Department of Earth and Environmental Sciences, University of Kentucky. Lexington KY, 40506 USA.
4. Centro Oceanográfico de Vigo, Instituto Español de Oceanografía, P.O. Box 1552, 36200 Vigo, Spain and School of Biological Sciences (Zoology), University of Aberdeen, Tillydrone Avenue, AB24 2TX Aberdeen, United Kingdom

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Corresponding author: angelguerra@iim.csic.es. Tel: + 34 986 214458; Fax: + 34 986 292762

ABSTRACT

Carbon and nitrogen isotope profiles constructed from the upper beaks of four giant squid *Architeuthis dux* provided time-integrated record of dietary information for individuals that inhabited Asturias (North Iberian Peninsula) and Namibian waters. $\delta^{15}N$ values ranged from 6.3 to 7.1 ‰ and $\delta\,^{13}C$ values ranged from -11.4 to -10.2. Nitrogen isotope profiles differed significantly between the four animals analyzed. These profiles differed in shape but all $\delta^{15}N$ increased along each profile with lowest values (but a higher rate of increase) around the rostral tip. The increase in $\delta\,^{13}C$ values was not as pronounced as for the $\delta^{15}N$ profiles. The changes in isotope compositions suggest that early in the life of the giant squid there is an ontogenetic shift in diet from smaller prey of relatively low trophic status to larger prey of higher trophic status. Fluctuations in $\delta\,^{13}C$ values observed near the rostral tip may be associated with a greater intrinsic variability in the carbon isotope composition of relatively small prey, and/or they may be attributed to transient migratory behaviour early in the life of individuals. The relative stability of the $\delta\,^{13}C$ profiles afterwards suggests that adult giant squids probably inhabit relative small and well-defined productive areas where food resources have a constant carbon isotope composition. The $\delta\,^{15}N$ and $\delta^{13}\,C$ values of giant squid beaks from North Iberian Peninsula and Namibia are distinct, reaching higher values in Namibia, which suggest that the foraging ecology or food webs are different for animals in these marine ecosystems.

INTRODUCTION

Stable isotope signatures of both hard and soft tissues of animals are very useful tools in ecological research (Jackson et al., 2007; Michener and Lajtha, 2007) that can be used to better understand the function and role of organisms in food webs (Gannes et al., 1997; Michener and Lajtha, 2007). Stable isotope signatures can also provide information about the geographic origin and/or migratory behaviour of animals because the isotopic composition of food and water can vary spatially in systematic and predictable ways (Graves et al., 2002; Michener and Lajtha, 2007).

Stable isotope analysis has been used in several previous studies on cephalopods. For example, the carbon- and oxygen-isotope composition of the cuttlebone of wild and cultivated specimens of the cuttlefish *Sepia officinalis* provides accurate estimates of seasonal changes in sea water temperature, which can be used to determine life history traits including life span, reproductive strategies, and growth rates (Guerra and Bettencourt, 1999). Stable isotope signatures of captive *Lolliguncula brevis* clearly distinguished between animals feeding on different diets (Stowasser *et al.*, 2006). Lukener *et al.*, (2008) analyzed the carbon and oxygen isotope composition of *Spirula spirula* shells from three major oceans indicates developmental changes paralleling depth distributions. Stable isotope may be especially useful for cryptic organisms like the giant squids in which direct gathering of ecological information is normally not possible, but where the chitinous mandibles (beaks) are available for analysis.

Squid beaks are hard structures composed of chitin-protein complexes (Hunt and Nixon, 1981), which resist digestion, and can accumulate in the stomachs of predators (Clarke, 1980). Due to the incremental accretion of non-reactive biological material they retain a molecular record of growth from early development to the time of death, which can be deciphered to better understand the ecology of these animals (Cherel and Hobson, 2005). Cherel and Hobson (2005) showed that

$\delta^{13}C$ and $\delta^{15}N$ values can differ in different tissues from the same individuals, reflecting differences in biochemical composition and turnover rate, with beaks slightly enriched in $\delta^{13}C$ but highly impoverished in $\delta^{15}N$ compared with lipid-free muscle tissues. Moreover, beaks from the same species showed a progressively increased increase in isotope ratios with size, which suggested a dietary shift from lower to higher trophic level during growth. In the same way, $\delta^{15}N$ values of various parts of a single beak increased from the rostrum to lateral walls and wings (Fig.1). They also showed that $\delta^{15}N$ values of various species living in slope waters of the subantarctic Kerguelen Islands (n=18 species) encompass almost three distinct trophic levels, with a continuum between crustacean- and fish-eaters and a distinctly higher trophic level occupied by the colossal squid *Mesonychoteuthis hamiltoni*. Conversely, $\delta^{13}C$ values demonstrated that the cephalopods occupied three different marine ecosystems, with 16 species living and developing in Kerguelen waters and two species migrating from either Antarctica (*Slosarczykovia circumantarctica*) or the subtropics (the giant squid *Architeuthis dux*).

Stable isotope analysis can reveal many aspects of the life cycle, behaviour and ecology of *Architeuthis* spp., which are not well understood even though these squid have received considerable attention during the last few years (González et al., 2002; Guerra et al., 2004, 2006; Kubodera and Mori, 2005). Most of the available information is derived from dead or stranded animals, and from their predators (Clarke 1996; Klages, 1996; Collins, 1998; Santos et al., 2002). Paralarvae and juvenile individuals are epi-mesopelagic animals that inhabit surface waters down to about 700 m depth, whereas the adults are meso-bathypelagic living close to the sea floor in deeper waters (to 1500 m) and particularly associated with submarine canyons (Guerra et al., 2005). The age, growth rate and life span of *Architeuthis* are still open questions (Guerra et al., 2006; Grist and Jackson, 2007). Little is known about the diet of adult giant squids, but data indicate that they mainly eat other cephalopods and crustaceans, as well as a large proportion of fish of different families according to the available prey in the area (Guerra et al. 2006 for a review). However, inferences from the gut contents are difficult to make based on the fact that squid tear their prey into small pieces using their chitinous beaks, which prevents identification of prey remains in many cases. Giant squid have roles as both predators and prey (Clarke, 1980; 1996), but the lack of information on their trophic ecology hinders a better understanding of their role in marine ecosystems

In this paper, we have analysed the stable isotope composition of nitrogen ($\delta^{15}N$) and carbon ($\delta^{13}C$) along beaks of individual specimens of *A. dux*, which provide a continuous record of the life history experienced for these animals. The beak is secreted by an epithelium which adds chitinous material of the posterior side of the hood and wings and on the lateral sides of the lateral wall. As growth proceeds, the secreting area expands so that the chitinous material overlaps the edges of the growing surface and this overlapping gives rise to microrings. These rings or minute steps have been used different time to estimate age and growth of the animals (Clarke, 1965; 1993; Raya and Hernández-González, 1998; Hernández-López et al., 2001).The material deposited in the rostral tip is the oldest one providing information when the animal was a paralarva, while the posterior edge of the hood provides information on the adult phase (Clarke, 1980; Pérez-Gándaras, 1983).

To our knowledge, this study is the first to measure the stable isotopes profile along a cephalopod beak, considering that a single beak can provide information about some bioecological features of the animal over the course of its life. due to the fact that its growth for layers periodically settled keeps this information. This approach is based on the hypothesis of an enrichment of beak nitrogen and carbon isotopes occur with growth.

The aims of this paper are to better understand the diet, habits and migratory behaviour for the species, using stable isotopes signatures recorded in the beak over the lifetime of four individual squid from Spain and Namibia, and to evaluate the degree of individual and geographic variation in the ontogenetic patterns observed.

MATERIAL AND METHODS

Specimens

We analysed three maturating adult females of *Architeuthis dux* Steenstrup 1857 (specimen numbers 1933, 1964 and 1963) captured by pair trawlers at *Carrandi* fishing ground (43º 52.54'/ 43º 52.75 N - 05º 06.30'/ 05º18.74 W, Western Asturian waters, North Spain); at depth between 260 and 570 m, and one immature female (1427) *Architeuthis aff. dux* fished by an otter trawler off Namibian waters (22º 56.25'/23º 05.18' S- 14º 28.29'/ 13º18.14 E, depth: 350-390 m, Table 1). The three Spanish animals were found stranded, complete and fresh. After collection, they were frozen at -20º C**.** The Namibian specimen was frozen on board at -40º C**.** Beaks were removed from defrosted carcasses in the laboratory at room temperature and preserved in ethanol 70 %.

Stable isotope methods

The upper mandible of each beak (Fig.1) was serially sampled every 1.43 mm from the rostral tip of the hood to the posterior edge of the hood. Material was collected for isotope analysis by cutting out 1x1 mm pieces using a surgical scissors. Subsamples (1 mg) were loaded into pre-cleaned tin cups and placed in an Elemental Analyzer (NC 2500, CE Instruments) attached to a continuous flow isotope ratio mass spectrometer (Delta plus XL, Finnigan) for carbon and nitrogen stable isotope analysis. Stable isotope ratios ($^{13}C/^{12}C$ and $^{15}N/^{14}N$) are reported in per mil (‰) units using δ notation relative to Vienna Pee Dee belemnite (carbon) international standard and air. An international certified standard (DORM-2) was analyzed periodically and was reproducible to better than ± 0.1 %o (1 σ SD n = 38) for both $\delta\,^{13}C$ and $\delta\,^{15}N$ values.

Statistics

Since each segment was approx. 1.5 mm in length, over the 70 mm length of the hood of the largest upper beak there were only 47 separate segments and the other series were shorter. All data series were tested for autocorrelation. Autocorrelation was observed for lags of 1 to 3 positions but partial autocorrelations were generally significant ($p<0.05$) only for lag 1 (nitrogen data) or lags 1 and 2 (carbon data).

The length of the data series differed between animals and for this reason we approximated an AR(1) or AR(2) structure by including the isotope ratios for the previous one or two segments as linear predictors. Using this approach, the isotope ratios for the first one or two segments in each series (starting at the rostral tip) are excluded from analysis since previous values are not defined.

Generalised Additive Models (GAM) were fitted to the isotopic ratio data, the ratio being the response variable and with position (segment number) treated as a continuous explanatory variable (fitted as a smooth curve) and animal as a categorical factor and the previous one (N) or two (C) isotopic ratios in the series as linear predictors. The isotope values were approximately normally distributed so a Gaussian GAM with identity link function was used. The “full model” for each isotope included separate smoothers for each animal for the position effect. The model for nitrogen was thus:

Y1 ~ 1 + s(Position, by = as.numeric (Animal_1427)) + s(Position, by = as.numeric (Animal_1932)) + s(Position, by = as.numeric (Animal_1963)) + s(Position, by = as.numeric (Animal_1964)) + as.factor(Animal) + Previous ratio.

For both isotopes, the full model was compared, using an F test, with models with (a) separate smoothers for the position effect for each country and country (rather than individual) as a categorical explanatory variable, (b) a combined smoother for all animals for the position effect, (c) without the previous isotope ratios as predictors. In all cases, model residuals were also checked for patterns. All models were fitted using BRODGAR software (www.brodgar.com) and procedures are based on those described in Zuur et al. (2007).

RESULTS

Figure 2 shows the $\delta^{15}N$ profile of the hood of the upper beak of the four *Architeuthis* specimens of this study. Each sample (Appendix 1) is marked by a symbol. In the axis of abscissas the distance is indicated from the rostral tip of hood, which represents the earliest stage of life, up to the posterior edge of the hood, which represents the adult stage of life. Figure 3 shows similar information for the carbon isotope profiles. It is clear that there is a general upward trend over time, consistent with an ontogenetic increase in trophic level. As would be expected, this trend is clearer in the nitrogen data. It is also apparent from the nitrogen data that the rate of increase in trophic level is higher in the early part of the life (as recorded nearer to the rostral tip, see Figure 2), although it should be borne in mind that this is based on the rate of change in the isotope ratios in relation to units of beak growth rather than units of time.

In the case of carbon, it is notable that there is some fluctuation in values close to the rostral tip and it also appears that the isotope ratio reaches an asymptote and starts to decrease towards the end of the animal’s life (Figure 3).

The full GAMs for both nitrogen and carbon isotopes performed better than any of the simply models and the residuals were free from important patterns (see Tables 2 and 3 for details). Thus, significant differences in isotope ratio trajectories are confirmed between animals and provide better models than treating animals from the two countries separately or grouping all four animals together. However, it can be seen from the signs of the effects of the categorical variable “animal” that the beak of the Namibian animal was more enriched in both C and N than any of the three Spanish animals. The fitted smoothers for the effect of position were generally non-linear, as shown by the degrees of freedom for the smoothers usually exceeding 1.0 (except for nitrogen in animal 1964). As expected from viewing the raw data, all smoothers indicated a generally upward trend in both C and N isotope ratios as position along the beak moved further from the rostral tip.

The consistent changes in the slopes of the relationships between isotope ratio and position along the beak are less evident from the fitted smoothers (not shown) than from the raw data (Figure 2 and 3), presumably because autocorrelation has been removed from the former.

DISCUSSION

Our δ^{15}N profiles verified our hypothesis of an enrichment of nitrogen isotope in the beak with growth, which may be interpreted as evidence of an ontogenetic dietary shift. The δ^{15}N values in the upper rostral tip correspond to the youngest phases of the giant squid life. They suggest that in early life, the squids fed at a lower trophic level than later in life. Data on gut contents from adult specimens suggest that the diet comprises mainly mesopelagic (200-1000 m depth) cephalopods and fishes (Deagle et al., 2005; see Guerra et al., 2006 for a review). However, no information exists about the feeding habits of *Architeuthis dux* paralarvae and juveniles. Nevertheless, an ontogenetic change in feeding habits, from paralarvae and early juveniles feeding on small planktonic and pelagic prey, low in the food web, to adults feeding on larger prey higher in the food web is well-documented in most short-lived (≥ 2 years) squids (Rodhouse and Nigmatullin, 1996; Boyle and Rodhouse, 2005).

We observed a 4.5-7.5 ‰ difference in δ^{15}N values between the rostral tip and the posterior edge of the hood of the four *Architeuthis* analysed. The range of this shift is three times larger that the found by Cherel and Hobson (2005) in medium and large beaks of the greater hooked squid *Moroteuthis ingens* (1.9 ‰), and also larger than the difference found between small and large beaks of *Kondakovia longimana* (4 ‰). Accordingly to DeNiro and Epstein (1978), Minigawa and Wada (1984) and Vanderklift and Ponsard (2003) a 4 ‰ shift in δ N value is equivalent to approximately one trophic level although if the enrichment from one trophic level to the next is lower, it could represent two or more trophic levels. Differences in δ N values reported by Cherel and Hobson (2005) were interpreted to represent a dietary shift mainly from crustaceans to mesopelagic fishes in *M. ingens*, and *K. longimana*.

The giant squid is distinguished from *M. ingens* and *K. longimana* by having higher δ^{15}N values. However, the δ^{15}N values found in the posterior edge of the hood of *A. dux* (9.9-13.4 ‰) are close to those of the large beaks of the colossal squid *Mesonychoteuhis hamiltonii* (Cherel and Hobson, 2005). This suggests that both giant and colossal squid are higher top predators in their respective ecosystems. It is notable that giant squids (*A. dux*) from North Iberian Peninsula and Namibia (present paper) and those from Kerguelen Islands (Cherel and Hobson, 2005) are segregated by their $\delta\ ^{15}$N values (5.3-13.5 *versus* 6.4-8.6) The differences in nitrogen isotopes values could demonstrate that these giant squids grew in different marine ecosystems.

As expected, we observed differences in the δ N value at different points analysed along the hood of the upper beak with an increase from the rostrum to the posterior edge, which suggest an ontogenetic shift in the feeding habits of *Architeuthis*. This finding is similar to the observed by Cherel and Hobson (2005) when comparing the values of δ N in different parts of the same beaks, with an increase from the rostrum to the lateral walls and a further increase from the lateral walls to the wings (Fig. 1). The material in the lateral walls and the wings of the beaks, as well as the posterior edge of the hood, is the last deposited and is therefore indicative of the feeding during the later period of the animal's life.

The four upper beaks analysed in the present paper showed the δ^{13} C values increased from -17.8 to -15.8 ‰ in specimens 1932, 1964 and 1963, and from -16.8 to -14.3 ‰ in 1427. These different values suggest that the three specimens from Asturian waters lived in different water mass to that occupied by the specimen from Namibia. On the other hand, the δ^{13} C values also increased over the life of the four specimens (Fig. 3). Although a gradual increase is expected with increasing trophic level, this difference could also result from movement between different foraging areas with distinct primary sources of carbon (DeNiro and Epstein, 1978; Hobson *et al.*, 1994; Cherel *et al.*, 2000). Fluctuations in the δ ^{13}C profiles near the rostral tip (Fig.3) may be associated with greater intrinsic variability in the carbon isotope composition of relatively small prey (Cabana and Rasmussen 1996) on which the giant squid depended early in life. On the other hand, the differences may be attributed to migratory behaviour early in the life of the squid.

Relative stability in the δ ^{13}C profiles approximately after 20 mm (Fig. 3) may be associated with a more constant and defined food resource later in life. The constancy of the carbon isotope values could also mean that adult giant squid live in a single area feeding on prey of similar carbon isotope composition. Sedentary behaviour would be consistent with the poor swimming abilities of *Architeuthis* (Roper & Boss 1982; Guerra et al., 2006). To maintain the high growth rate characteristic of these animals (Grist and Jackson, 2007) given a relatively low locomotion efficiency and relatively low metabolism (Seibel et al., 2000), implies *Architeuthis* must live in areas of relatively high productivity. The locations of *Architeuthis* catches show a high concordance with relatively close abyssal depths, submarine canyons, and water masses characterized by high productivity (Guerra et al., 2005).

As observed in δ N values, giant squids from North Iberian Peninsula and Namibia (present paper) and those from Kerguelen Islands (Cherel and Hobson, 2005) are also segregated by their δC values (-13.9 to -17.8 *versus* -17.0 to -17.8 ‰). This difference in carbon isotope values strongly reinforces the indication that these giant squids grew in distinct marine ecosystems.

CONCLUSION

Stable isotope analyses of a unique beak provide useful information about the life history of otherwise inaccessible creatures such as the giant squid. The nitrogen isotope composition along the hood of the upper beak was especially useful in revealing ontogenetic shifts in the feeding habits of *Architetuhis dux* from diets rich in small prey early in life to larger prey latter in life. The carbon isotope record could indicate that paralarvae and juveniles of *A. dux* have an intense migratory behaviour, feeding on distinct planktonic primary sources of carbon, whereas the adult giant squids probably stay in a relatively small and well defined area where the food resources have a relatively constant carbon isotope composition, and that adult *Architeuthis* must live in areas of relatively high productivity. The differences in nitrogen and carbon isotopes ratios of specimens from different geographic areas seem to demonstrate that the giant squids grew in marine ecosystems of different characteristics.

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Table 1. *Architeuthis* specimens analyzed in this study. S. No: specimen number at Ecobiomar Research Group's Archive (Instituto de Investigaciones Marinas, Consejo Superior de Investigaciones Científicas, Vigo) ML: mantle length (cm); BW: body weight (Kg); S: sex; F: female; MS: Maturity Stage; Ma: Maturing; Im: Immature; UHL (mm): Upper Hood Length.

S. No	**Locality**	**ML**	**BW**	**S**	**MS**	**UHL**	**Comments and sources**
1932	Carrandi	150	104	F	Ma.	60.4	Fresh stranded. Date: 23.10.2001. González et al., 2002; Guerra et al., 2004
1964	Carrandi	152	105	F	Ma.	59.6	Fresh stranded. Date: 15.09.2003. González et al., 2002. Guerra et al., 2004
1963	Carrandi	153	140	F	Ma.	70.2	Fresh stranded. Date: 13.09.2003. González et al., 2002; Guerra et al., 2004
1427	Namibia	105	47	F	Im.	56.2	Caught by trawler. Date: 09.02.1990.

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Table 2. GAM results for nitrogen isotope ratios in the hood (N=163). The explanatory variables were animal number (using animal 1427 as the reference animal) and position (nested within animal number). "S(X)" indicates a smoothing function with X degrees of freedom. An AR(1) time series structure was approximated by using the isotope ratio from the previous position on the beak as a linear predictor.

Parameter	**Value**	**Test statistic**	**Value**	**Probability**
Previous ratio	0.510	T	13.50	<0.0001
Animal 1932	-1.041	T	-12.38	<0.0001
Animal 1963	-0.788	T	-11.32	<0.0001
Animal 1964	-0.851	T	-11.57	<0.0001
Position: animal 1427	S(3.26)	F	15.73	<0.0001
Position: animal 1932	S(4.88)	F	8.01	<0.0001
Position: animal 1963	S(6.28)	F	12.68	<0.0001
Position: animal 1964	S(1.00)	F	58.45	<0.0001
R-sq (adj.)	0.983			

Table 3. GAM results for carbon isotope ratios in the hood (N=159). The explanatory variables were animal number (using animal 1427 as the reference animal) and position (nested within animal number). "S(X)" indicates a smoothing function with X degrees of freedom. An AR(2) time series structure was approximated by using the isotope ratios from the previous two positions on the beak as linear predictors.

Parameter	**Value**	**Test statistic**	**Value**	**Probability**
Previous ratio	-0.143	T	-2.14	0.0343
Lag 2 ratio	0.222	T	4.04	<0.0001
Animal 1932	-1.090	T	-8.23	<0.0001
Animal 1963	-1.357	T	-8.63	<0.0001
Animal 1964	-0.824	T	-7.51	<0.0001
Position: animal 1427	S(7.88)	F	15.93	<0.0001
Position: animal 1932	S(7.07)	F	13.42	<0.0001
Position: animal 1963	S(7.79)	F	11.54	<0.0001
Position: animal 1964	S(4.96)	F	6.12	<0.0001
R-sq (adj.)	0.98			

Figure captions:

Figure 1. Upper beak of *Architeuthis*. UHL: Upper hood length (Clarke, 1980).

Figure 2. Nitrogen isotope profiles along the hood of the upper beak of females *Architeuthis*. Specimen numbers as in Table 1; D (mm): distance from the rostral tip to the posterior edge of the hood.

Figure 3. Carbon isotope profiles along the hood of the upper beak of females *Architeuthis*. Specimen numbers as in Table 1; D (mm): distance from the rostral tip to the posterior edge of the hood.

Figure 1

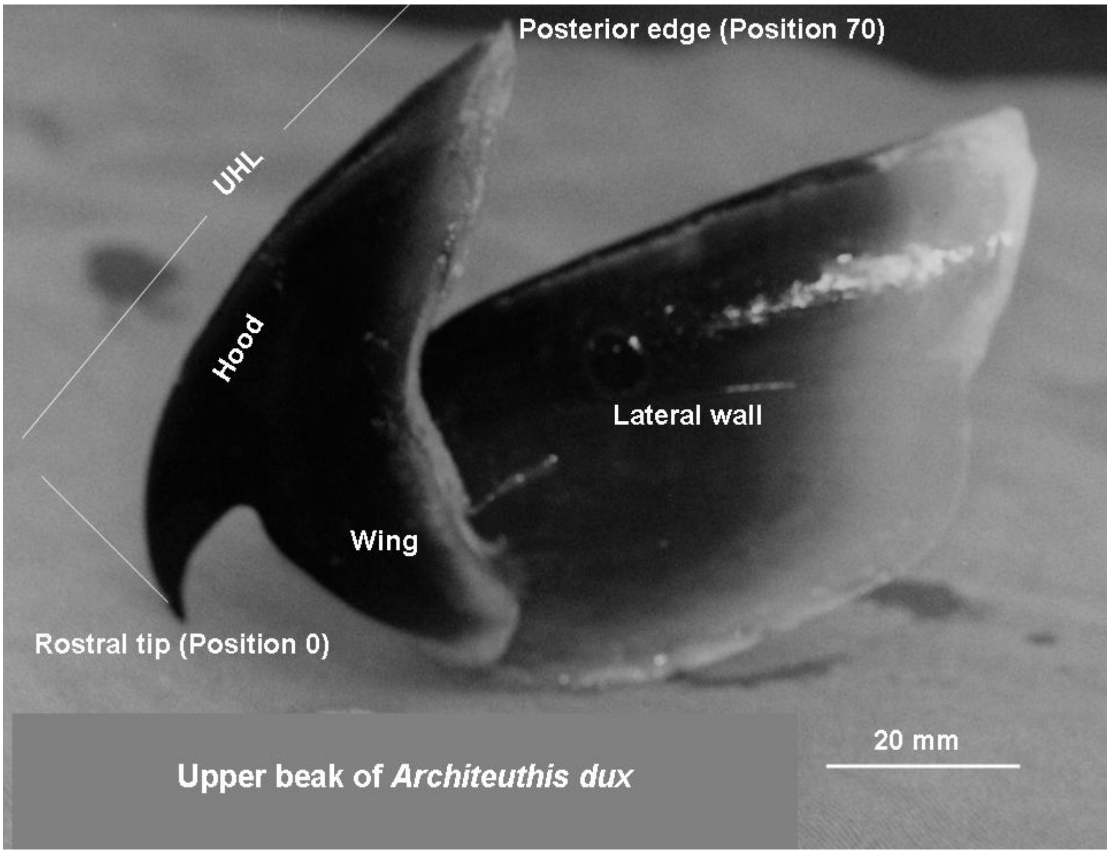

Posterior edge (Position 70)
UHL
Hood
Lateral wall
Wing
Rostral tip (Position 0)
20 mm
Upper beak of *Architeuthis dux*

Figure 2

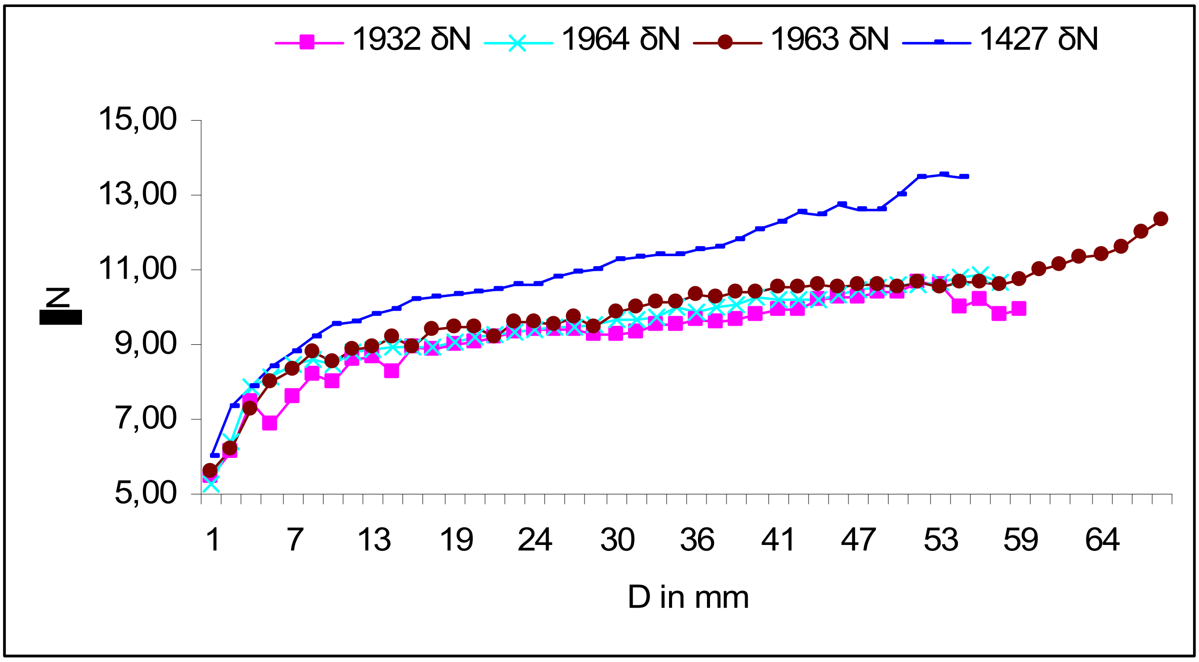
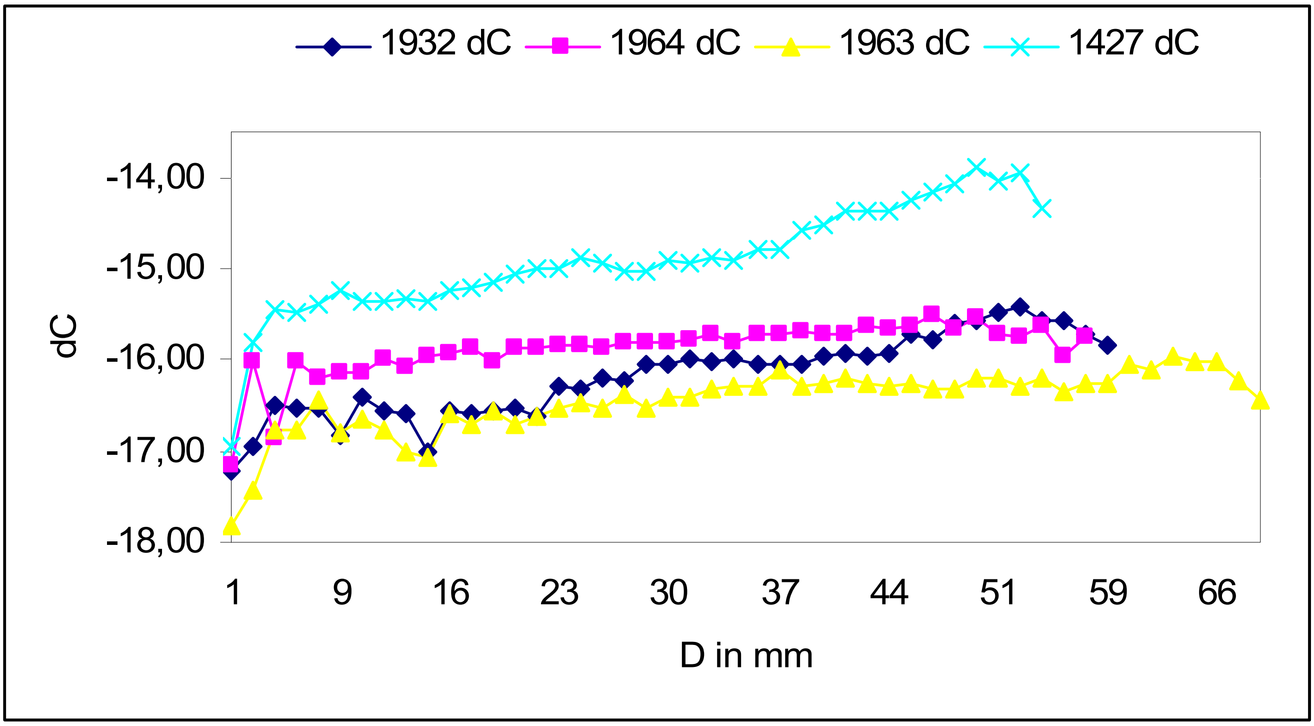

1932 δN
1964 δN
1963 δN
1427 δN
15,00
13,00
11,00
9,00
7,00
5,00
N
1
7
13
19
24
30
36
41
47
53
59
64
D in mm

Figure 3


1932 dC
1964 dC
1963 dC
1427 dC
-14,00
-15,00
-16,00
-17,00
-18,00
dC
1
9
16
23
30
37
44
51
59
66
D in mm

Appendix 1. Raw data of $\delta^{15}N$ and $\delta^{13}C$ values in the hood of the upper beaks of *Architeuthis* from Asturias (North Spain, specimen numbers (S. No) 1932, 1964 and 1963) and Namibian waters (1427). D: distance (in mm) from the rostral tip to the posterior edge of the upper beak where each sample was taken.

S.No	**1932**	**1932**	**1964**	**1964**	**1963**	**1963**	**1427**	**1427**
D	**δN**	**δC**	**δN**	**δC**	**δN**	**δC**	**δN**	**δC**
1,43	5,48	-17,21	5,27	-17,16	5,61	-17,81	5,98	-16,94
2,86	6,12	-16,96	6,38	-16,03	6,22	-17,43	7,35	-15,81
4,29	7,49	-16,51	7,87	-16,86	7,24	-16,76	7,84	-15,46
5,72	6,84	-16,54	8,11	-16,02	7,99	-16,76	8,39	-15,49
7,15	7,61	-16,52	8,46	-16,20	8,33	-16,45	8,77	-15,40
8,58	8,19	-16,82	8,57	-16,14	8,82	-16,79	9,17	-15,23
10,00	8,01	-16,40	8,49	-16,15	8,53	-16,64	9,53	-15,36
11,43	8,62	-16,56	8,78	-15,99	8,84	-16,78	9,63	-15,35
12,86	8,65	-16,59	8,86	-16,08	8,95	-17,01	9,82	-15,33
14,29	8,28	-17,02	8,91	-15,95	9,21	-17,07	9,96	-15,36
15,72	8,95	-16,56	8,91	-15,93	8,93	-16,58	10,19	-15,23
17,15	8,88	-16,60	8,94	-15,88	9,41	-16,70	10,28	-15,20
18,58	9,03	-16,55	9,08	-16,02	9,44	-16,55	10,35	-15,14
20,01	9,09	-16,52	9,23	-15,86	9,44	-16,71	10,39	-15,07
21,44	9,17	-16,62	9,25	-15,88	9,23	-16,62	10,49	-15,01
22,87	9,31	-16,28	9,33	-15,84	9,62	-16,54	10,57	-15,01
24,30	9,43	-16,31	9,38	-15,83	9,57	-16,46	10,61	-14,89
25,73	9,38	-16,21	9,50	-15,86	9,54	-16,53	10,79	-14,95
27,16	9,39	-16,24	9,45	-15,80	9,74	-16,39	10,92	-15,02
28,59	9,29	-16,05	9,55	-15,82	9,47	-16,52	11,03	-15,04
30,02	9,27	-16,05	9,67	-15,80	9,90	-16,40	11,25	-14,90
31,45	9,36	-15,99	9,69	-15,79	9,97	-16,42	11,33	-14,94
32,88	9,52	-16,03	9,75	-15,72	10,16	-16,32	11,40	-14,89
34,31	9,51	-15,98	10,01	-15,81	10,15	-16,29	11,42	-14,90
35,74	9,67	-16,05	9,87	-15,71	10,31	-16,30	11,51	-14,79
37,17	9,62	-16,05						-14,79
38,60	9,66	-16,05	10,06	-15,70	10,39	-16,29	11,81	-14,58
40,03	9,78	-15,95	10,24	-15,72	10,42	-16,27	12,10	-14,52
41,46	9,95	-15,92	10,20	-15,72	10,51	-16,20	12,28	-14,37
42,89	9,96	-15,96	10,21	-15,64	10,54	-16,27	12,54	-14,38
44,32	10,17	-15,94	10,23	-15,66	10,57	-16,29	12,50	-14,37
45,75	10,26	-15,73	10,34	-15,64	10,53	-16,26	12,73	-14,26
47,18	10,28	-15,78	10,44	-15,50	10,62	-16,32	12,59	-14,16
48,61	10,38	-15,59	10,52	-15,66	10,61	-16,31	12,63	-14,07
50,04	10,37	-15,57	10,58	-15,55	10,55	-16,20	12,97	-13,88
51,47	10,69	-15,47	10,63	-15,72	10,68	-16,19	13,45	-14,05
52,90	10,59	-15,41	10,69	-15,75	10,56	-16,29	13,54	-13,94
54,33	9,98	-15,56	10,78	-15,63	10,65	-16,20	13,45	-14,34
55,76	10,17	-15,58	10,84	-15,95	10,64	-16,35		
57,19	9,81	-15,73	10,68	-15,76	10,61	-16,26		
58,62	9,96	-15,83			10,76	-16,25		
60,05					10,99	-16,06		
61,48					11,14	-16,11		
62,91					11,34	-15,97		
64,34					11,41	-16,03		
65,77					11,57	-16,01		
67,20					12,02	-16,22		
68,63					12,32	-16,43		