

Jack of all prey, master of some: Influence of habitat on the feeding ecology of a diving marine predator

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Abstract Marine species occupy broad geographical ranges that encompass varied habitats. Accordingly, resource availability is likely to differ across a species range and, in-turn, this may influence the degree of dietary specialization. Gentoo penguins *Pygoscelis papua* are generalist predators occupying a range of habitats with a large breeding range extending from Antarctica to temperate environments. Using the most extensive stomach content data set on gentoo penguins this study investigated their feeding ecology at the Falkland Islands (52°S, 59.5°W), the world's largest population. Sampling occurred in

consecutive breeding seasons (2011–2013), across multiple foraging habitats utilizing stomach content data and carbon and nitrogen stable isotope values of feathers. The first species specific description of diet at this scale for the Falklands revealed six key prey items for the birds: rock cod (*Patagonotothen* spp.), lobster krill (*Munida* spp.), Falkland herring (*Sprattus fuegensis*), Patagonian squid (*Doryteuthis gahi*), juvenile fish (likely all nototheniids), and southern blue whiting (*Micromesistius australis*). Niche width, relating to both stomach content and stable isotope data related to the surrounding bathymetry. Birds from colonies close to gently sloping, shallow waters, fed primarily on benthic prey and had larger niche widths. The opposite was observed at a colony surrounded by steeply sloping, deeper waters. Therefore, gentoo penguins at the population level at the Falklands are indeed generalists, however, at individual colonies some specialization occurred to take advantage of locally available prey, resulting in these birds being classified as Type B generalists. Hence, future studies must account for this intra-colony variation when assessing for factors such as inter-specific competition or overlap with anthropogenic activities.

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Introduction

Conspecifics occupying varied foraging habitats may have different dietary niche widths owing to their different ecological settings (Rosenzweig 2007; Newsome et al. 2015; Corman et al. 2016). Therefore, species that are considered to be generalists may in fact consist of populations that are specialized at a local scale (Jaeger et al. 2010; Matich et al. 2011). Where the term specialization designates the relative width of an individual's diet compared to that of the population (Bolnick et al. 2003). This makes knowledge of

whether a generalist species consists of individuals that all utilize a diverse array of prey items (Type A) or subsets of individuals that utilize specific prey items (Type B) essential to acquire, so as to understand and predict the capacity of populations to adapt to global and environmental changes (Van Valen 1965; Bearhop et al. 2004). Ultimately, differences may occur due to complex interactions between predator and prey which can arise from environmental heterogeneity (Bolnick et al. 2003). For seabirds in particular, only 40 species (11.7% of the total extant species) have exhibited some type of individual specialization in foraging and/or feeding strategies, but this proportion likely reflects existing studies (Ceia and Ramos 2015).

To gain insight toward the dietary niche width of an organism one can utilize a combination of direct and indirect methods. Using a combination has added value as it allows insight into dietary specialization over varying temporal scales (Ceia et al. 2012; Thomson et al. 2012). Specifically, the direct method of stomach content analysis, enables identification of prey items to species level and one can determine the mass and size of prey items consumed (Jobling and Breiby 1986; Granadeiro and Silva 2000; Barrett et al. 2007). However, stomach sampling represents a snap shot of ingested prey items (the most recent meal), is invasive and logistically challenging for sustained sampling over extended time periods (Duffy and Jackson 1986; Karnovsky et al. 2012). Accordingly, one can also utilize an indirect method, stable isotope analysis, to investigate carbon and nitrogen isotopic values ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$). These values indicate the isotopic niche of a consumer, which can be used to infer its foraging area and trophic position in a given geographic area, respectively (Hobson et al. 1994; Cherel and Hobson 2007; Jaeger et al. 2010). For birds, these values are often measured in feathers as storage and collection is simplified compared to the more intensive requirements for blood sampling (Inger and Bearhop 2008; Bond and Jones 2009). Additionally, feathers are metabolically inert and therefore reflect the trophic ecology of individual birds at the time of deposition (Mizutani et al. 1990; Cherel et al. 2000; Polito et al. 2011b).

The gentoo penguin, *Pygoscelis papua*, is often considered a generalist and opportunist marine predator, with diet being varied and reflecting local prey availability (Clausen et al. 2005). During the breeding season adults seldom travel further than 30 km's and typically remain within the confines of the continental shelf (Trivelpiece et al. 1986; Wilson et al. 1998; Miller et al. 2009). Although, in rare instances have been recorded to travel as far as 46 km (Lescroël and Bost 2005). Hence, gentoo penguin diet is influenced by available habitat within close proximity to breeding colonies (Hindell 1989; Lescroël et al. 2004; Miller et al. 2010). In these studies, benthic prey items were typically associated with a gently sloping, shallower

sea floor in the vicinity of the colony, whereas pelagic feeding was more typical in deep surrounding waters.

The Falkland Islands present a unique opportunity for multi-colony dietary investigation across a wide range of habitat features. This archipelago now hosts the largest population of gentoo penguins with 34% of the global population, $\approx 132,000$ breeding pairs (Lynch 2012; Baylis et al. 2013). Therefore, understanding factors influencing the foraging ecology of this population is of significance in managing and understanding the species as whole. Dietary studies at the Falkland Islands are currently limited to a description of stomach content analysis at the level of broad taxonomic composition (Pütz et al. 2001, Clausen and Pütz 2002), or more detailed studies relating to a single year (Clausen et al. 2005) or colony (Handley et al. 2015). A detailed, spatially extensive, dietary study of the gentoo penguin at the Falkland Islands is therefore timely. It will also facilitate our understanding of conservation needs for these birds, in light of current and proposed anthropogenic activities for the Islands (Augé et al. 2015).

Here, we use the most extensive dataset that integrates stomach content analysis (detailed dietary analysis during the breeding period), complemented with feathers' stable isotopes, a proxy for foraging distribution and trophic interactions relating to the adult pre-moult diet of gentoo penguins. We selected colonies where the surrounding habitat was either shallow with a gently sloping seabed or deep with a steeply sloping seabed in order to determine how these habitats might influence diet. Specifically, the aims of the study were to (1) give detailed description of dietary items at a large spatial scale across the Falkland Islands, (2) assess temporal (between breeding stages) and spatial (across colonies, controlling for breeding stage) variation in the diet during the breeding period and (3) compare dietary and isotopic measures of gentoo penguin trophic niche width and specialisation in the context of different habitats. Through understanding each of these facets we address the degree of gentoo penguin dietary specialization at the Falkland Islands.

Methods

Study sites

Fieldwork occurred at the Falkland Islands during the austral summers of 2011/12, 2012/13 and 2013/14 hereafter referred to as 2011, 2012, 2013 respectively. Birds were sampled from four of the 75 breeding colonies: Steeple Jason Island (51.0375°S, 61.2097°W), Cow Bay (51.4288°S, 57.8703°W) Bull Roads (52.3096°S, 59.3896°W) and Bull Point (52.3478°S, 59.3287°W)

(Fig. 1), each with approximately 3710, 1821, 1236, 3400 breeding pairs, respectively (Stanworth 2013).

Each of these colonies is surrounded by a unique bathymetry and exposure to the ocean, particularly at Steeple Jason Island. The island lies between a steeply sloping seabed that rapidly drops down to 100 m whereas the Cow Bay and Bull Point colonies follow on to gently sloping seabeds facing the open ocean, while the Bull Roads colony is in a sheltered, shallow bay.

Sample collection

Stomach contents were collected from breeding adults during the incubation (I), guard (G) and crèche (C) stages. These occur during early November, late November/early December and January, respectively. Sampling effort varied spatiotemporally owing to logistical constraints. The most samples were collected during the guard period of 2011 from three colonies. Then, in the crèche period of 2012, we were able to collect samples from all four colonies. Ultimately, between 8 and 31 birds were sampled per colony in a given period and year (Supplementary material, Table S1). Where, a single sampling period refers to all birds sampled from a specific site in a given breeding stage and season. Methods associated with collection and sorting of stomach contents are detailed in Handley et al. (2015). Briefly, stomach lavage followed Wilson (1984) and three white breast feathers were also plucked from each bird for subsequent stable isotope analysis. As gentoo penguins molt synchronously at the Falkland Islands, the isotopic values of these feathers would be derived from dietary items consumed during the pre-moult diet period, which occurs towards the end of February. A random

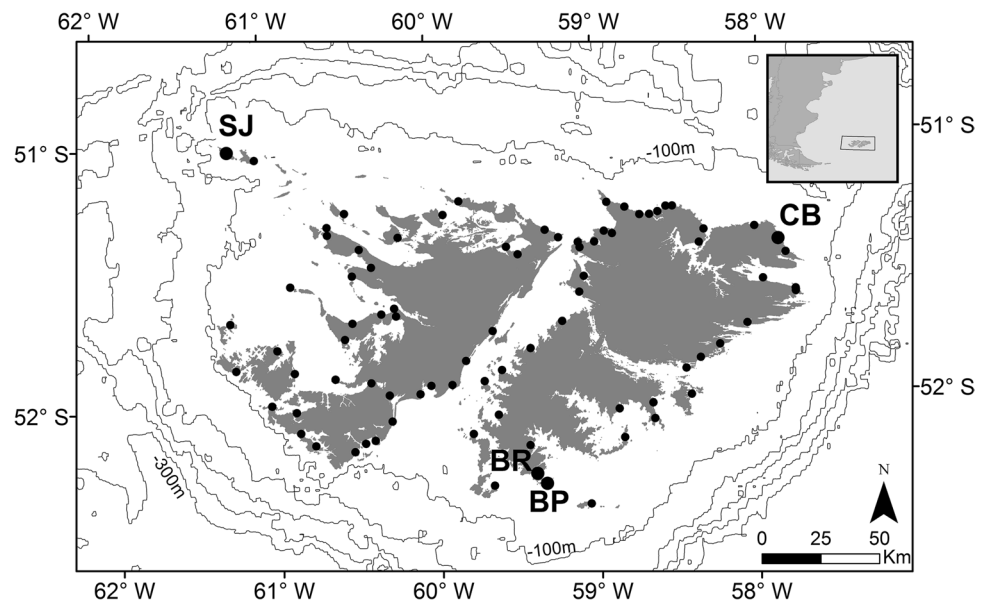
sub-sample of birds had feathers chosen for final processing and in some instances, we also bolstered sample size by using feathers plucked from birds involved in other ongoing studies. We adopted this pragmatic approach to achieve roughly equal sample sizes of 20 birds per sampling period.

Laboratory analyses

We gathered sagittal otoliths, cephalopod beaks, crustacean carapaces, or other hard part remains and identified these to the lowest possible taxonomic level by comparing them with reference collections maintained by the Falkland Islands Fisheries Department or published reference material (Clarke 1986; Xavier and Chérel 2009). Length and reconstituted mass of samples were calculated from non-eroded hard part remains (Van Heezik and Seddon 1989; Clausen et al. 2005) based on morphometric equations for each species following reference material or from regression equations developed during the study (Table S2). Wet mass was used in cases where whole prey could be identified, but no reference equation existed or could be developed (Clausen and Pütz 2003).

Regarding stable isotope analysis, two feathers from each bird were cleaned of contaminants and homogenized, in order to obtain an average value, following Connan et al. (2016). A sub sample of homogenized feather material weighing approximately 0.5 mg was analyzed for carbon and nitrogen stable isotope ratios ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$) via combustion in a Flash 2000 organic elemental analyzer with the gases passed to a Delta V Plus isotope ratio mass spectrometer (IRMS) via a ConFlo IV gas control unit (Thermo Scientific, Bremen, Germany). All samples were processed at the Stable Light Isotope Unit at the University of Cape

Fig. 1 Gentoo penguin colonies of the Falkland Islands (*small dots*) and the four study colonies: Steeple Jason (SJ), Cow Bay (CB), Bull Roads (BR) and Bull Point (BP). Isobaths (*faint lines*) are shown at 50 m intervals from 100 to 300 m depth



Town, South Africa. Results are presented in δ notation in per mil units (‰), based on the following equation:

$$\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 1000$$

where X is ^{13}C or ^{15}N and R is the corresponding ratio $^{13}\text{C}/^{12}\text{C}$ or $^{15}\text{N}/^{14}\text{N}$. The R_{standard} values were based on the Vienna PeeDee Belemnite (VPBD) for $\delta^{13}\text{C}$ and atmospheric nitrogen for $\delta^{15}\text{N}$. Replicate measurements of internal laboratory standards (Merck gel: $\delta^{13}\text{C} = -20.1\text{‰}$, $\delta^{15}\text{N} = +7.5\text{‰}$; seal bone $\delta^{13}\text{C} = -12.0\text{‰}$, $\delta^{15}\text{N} = +15.8\text{‰}$; valine $\delta^{13}\text{C} = -26.8\text{‰}$, $\delta^{15}\text{N} = +12.1\text{‰}$) indicated measurement errors $<0.2\text{‰}$ and $<0.1\text{‰}$ for stable-carbon and nitrogen isotope measurements, respectively.

Data analysis and statistics

Data analyses were performed using R version 3.2.1 (R Development Core Team 2015). Univariate normality and homoscedasticity were tested for via the Shapiro–Wilks and Bartlett’s test respectively. Multivariate analogues used were the multivariate Shapiro–Wilks test (package: `mvnormtest`, function: `m.shapirotest`) and the multivariate Levene’s test (package: `Vegan`, function: `betadisper`). In the case of linear or generalized linear models, model validation was performed via assessment for normal distribution of residuals, equality of variance and that no excessively influential observations were present (Zuur et al. 2009). Means with standard deviations are presented, and significance was assumed at $p < 0.05$ unless otherwise stated.

For stomach content data, we identified important prey items using the percentage index of relative importance (%IRI) as this facilitates comparison across sampling periods (Pinkas et al. 1971; Cortes 1997; Huin 2005). The IRI is an integrative metric that accounts for percentage mass, numerical abundance and frequency of occurrence. Analyses could then be conducted using the most important prey species (those with % IRI > 1). As the IRI relies on the summed information of each sampling period, yielding no variation in the results, percentage mass (%M) was chosen to compare prey items at the level of the sampling unit (each penguin), as this favours samples with varying prey size (Duffy and Jackson 1986; Ratcliffe and Trathan 2011). Sample size sufficiency was assessed through prey species accumulation curves and the Chao estimator (Chao 1987).

Comparisons were made between breeding stages for each colony in each season, then across different colonies while accounting for breeding stage and season. Differences in species composition were visually assessed with ordination via nonmetric multidimensional scaling (nMDS) (package: `Vegan`, function: `metaMDS` with

`autotransform = “F”`) using the %M data based on the major prey items across all sampling periods. Statistical differences were determined via the ADONIS test (package: `Vegan`, function: `adonis` with `distance = “bray”`).

Two dietary niche metrics were calculated for each sampling period (package: `RInSp`) (Zaccarelli et al. 2013). Firstly, the trophic niche width (TNW) was quantified using the Shannon–Weaver diversity index, following Roughgarden (1979). A value of 0 is scored when the entire population consumes a single prey category, increasing with both the number of prey species consumed and the evenness with which they are used. Secondly, diet variation among individuals was determined via the degree of individual diet specialisation (V), where a value of ‘0’ indicates that individuals use the same resources in the same proportions, and ‘1’ where individuals all rely on entirely different resources. The relationship between TNW and V was then assessed using a linear model.

Generalized linear models (GLMs) were used to compare $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values across sampling periods (family = `Gaussian`, link = `identity`), followed by the Tukey post-hoc test for pairwise comparisons (package: `multcomp`, function: `glht`). Prior to calculating measures regarding the isotopic niche space utilized by the penguins, data were scaled between 0 and 1 according to Cucherousset and Villéger (2015). This facilitates the spatial comparison when basal resources may differ and weights each isotope equally. The core isotopic niches across different sampling periods were investigated through the Stable Isotope Bayesian Ellipse package in R (SIBER) (Jackson et al. 2011). Differences in core niche width and overlap were explored by using the standard ellipse areas corrected for small sample size ($n < 30$), SEAc, which represents about 40% of the data. The Bayesian estimate of the standard ellipse area (SEA_B) was used to compare niche widths between groups in a probabilistic manner based on the size of simulated ellipse areas and their estimated posterior distributions (iterations = 10,000). Furthermore 95, 75 and 50% credibility intervals were assessed via density plots. The degree of overlap in these intervals is indicative of the degree of similarity in isotopic niche width between groups.

The methods outlined in Jackson et al. (2011) were developed to facilitate comparison between groups, particularly when there was varying sample size (e.g.: Connan et al. 2014; Kiszka et al. 2015; Polito et al. 2015). More recently, Cucherousset and Villéger (2015) proposed using complementary methods, which, include comparing the convex hull area, the entire space occupied by organisms in isotopic space (isotopic richness, *IRic*), something advised against by Jackson et al. (2011). However, Cucherousset and Villéger (2015) introduced a bootstrapping

approach to account for the varying sample size. They recognised the importance of investigating differences in convex hull area as this fully integrates the importance of organisms located at the edges of the isotopic niche. Therefore, the complementary metrics, isotopic richness, isotopic similarity (*ISim*) and isotopic nestedness (*INes*) were calculated. The bootstrapping approach was utilized when varying sample size was present (iterations = 4000). To account for the distribution of organisms within the isotopic niche space, isotopic divergence (*IDiv*), isotopic dispersion (*IDis*), isotopic evenness (*IEve*) and isotopic uniqueness (*IUni*) were also calculated. These metrics are briefly outlined in table S3, and were all calculated for scaled data.

Inferring diet: stable isotope mixing model

We attempted to quantitatively assess the contribution of different prey sources towards gentoo penguin diet using the SIAR Bayesian stable isotope mixing model (Parnell et al. 2010). A species and tissue specific discrimination factor of $1.3 \pm 0.5\%$ for $\delta^{13}\text{C}$ and $3.5 \pm 0.4\%$ for $\delta^{15}\text{N}$ were used (Polito et al. 2011a). Prey samples were dried and ground to a fine powder before lipid removal using cyclohexane (Chouvelon et al. 2011). Any prey item which still had a bulk C:N ratio >3.5 was subjected to $\delta^{13}\text{C}$ correction as outlined in Post et al. (2007). Despite using 17 prey items kindly sourced through research cruises undertaken by the Falkland Islands Fisheries Department, and reducing the number of sources to three (Phillips et al. 2005), the validity of the model was not suitable after using a Monte Carlo simulation approach (1500 iterations) to test the goodness-of-fit of the data to the model using simulated mixing polygons (Smith et al. 2013). See supplementary material for further discussion (Table S4 and Fig. S1).

Results

Stomach content analysis

A total of 13,197 individual prey items were identified over three seasons across the four study colonies. This included 8669 crustaceans, 4650 fish, 587 cephalopods and 11 other items from 237 penguins, comprising a total of 37 prey items (Table S5). A total of 19 of the 37 prey items were identified to species level and six of these contributed $>1\%$ IRI across all sampling periods combined (Table 1), the sizes of which are presented in the supplementary material, table S6. For each individual sampling period, prey species accumulation curves plateaued and there was zero deviation about the Chao estimator, as expected once a species is found in more than two individuals (Fig. S2).

Intra-annual variation in diet during the breeding period

During different stages of the 2011 and 2012 breeding seasons, significant differences in diet were evident in one of three, and three of four comparisons, respectively (Table 2, Fig. S3 (A–G)). In 2011, this difference was between the incubation and guard stage at Steeple Jason when birds consumed mainly Falkland herring (*Sprattus fuegensis*) compared to rock cod (*Patagonotothen* spp.) (Fig. S3 (A)). At the other two colonies during the 2011 breeding season birds consumed equally large proportions of rock cod then Patagonian squid (*Doryteuthis gahi*), or rock cod then lobster krill (*Munida* spp.) at Cow Bay (Fig. S3 (B)) and Bull Point (Fig. S3 (C)), respectively.

In 2012 significant differences occurred for all comparisons between the guard and crèche stages at Cow Bay, Bull Point and Bull Roads. At Steeple Jason birds fed equally on large proportions of Falkland herring during the incubation and crèche stages (Fig. S3 (D)). Differences were largely

Table 1 Main prey species identified from stomach content analysis of gentoo penguins at the Falkland Islands as indicated by having the percentage index of relative importance (%IRI) >1

Scientific name	Common name	Code	% IRI	Habitat	Source
Cephalopod					
<i>Doryteuthis gahi</i>	Patagonian squid	LOL	9.87	Benthic	Arkhipkin et al. (2013)
Crustacean					
<i>Munida</i> spp	Lobster krill	MUN	25.48	Benthic	Tapella and Lovrich (2006)
Fish					
<i>Patagonotothen</i> spp	Rock cod	PATA	46.55	Benthic	Brickle et al. (2006)
<i>Sprattus fuegensis</i>	Falkland herring	SAR	11.44	Pelagic	Zenteno et al. (2015)
Fish (juvenile)	Juvenile fish	JF	2.66	NA	NA
<i>Micromesistius australis</i>	Southern blue whiting	BLU	1.02	Pelagic	Brickle et al. (2009)
Others			2.98		

Typical habitat of prey items was determined from various sources

NA Data or source not available

Table 2 Results of ADONIS tests for comparison of major prey items between different stages of the breeding period (incubation (I), guard (G) and crèche (C)) and across colonies for gentoo penguins at the Falkland Islands

Season	Colony(ies)	Breeding stage(s)	<i>F</i> . model	<i>R</i> ²	<i>p</i> value	Fig.
2011	SJ	I vs. G	$F_{(1,38)} = 59.90$	0.61	0.001	S3 (A)
	CB	G vs. C	$F_{(1,38)} = 2.00$	0.05	0.092	S3 (B)
	BP	G vs. C	$F_{(1,37)} = 1.69$	0.04	0.185	S3 (C)
2012	SJ	I vs. C	$F_{(1,19)} = 0.35$	0.02	0.856	S3 (D)
	CB	G vs. C	$F_{(1,19)} = 11.55$	0.38	0.001	S3 (E)
	BP	G vs. C	$F_{(1,18)} = 3.87$	0.18	0.026	S3 (F)
	BR	G vs. C	$F_{(1,18)} = 5.24$	0.23	0.012	S3 (G)
2011	SJ, CB, BP	G	$F_{(2,82)} = 10.45$	0.2	0.001	S4 (A)
	CB, BP	C	$F_{(1,16)} = 2.06$	0.11	0.114	S4 (B)
2012	CB, BP, BR	G	$F_{(2,27)} = 3.35$	0.2	0.018	S4 (C)
	SJ, CB, BP, BR	C	$F_{(3,37)} = 8.68$	0.41	0.001	S4 (D)
2013	CB, BR	G	$F_{(1,34)} = 28.88$	0.46	0.001	S4 (E)

Samples were analysed for three seasons, 2011, 2012 and 2013 from four different colonies: Bull Point (BP), Bull Roads (BR), Cow Bay (CB) and Steeple Jason (SJ). Associated nMDS ordination and stacked bar plots can be found in supplementary material figures S3: A–G, S4: A–E

Significant differences are displayed in bold

due to different proportions of rock cod vs. Falkland herring, rock cod and Patagonian squid vs. lobster krill and juvenile fish, and, rock cod and Patagonian squid vs. lobster krill being consumed at Cow Bay (Fig. S3 (E)), Bull Point (Fig. S3 (F)) and Bull Roads (Fig. S3 (G)), respectively.

Variation in diet across colonies

In four of five inter-colony comparisons, significant differences in diet were observed (Table 2, Fig. S4 (A–E)). These occurred during all three years in the guard stages and in one of the two crèche stages investigated. Bull Point birds consumed significantly more lobster krill compared to rock cod dominating at Cow Bay and Steeple Jason during the guard stage of 2011 (Fig. S4 (A)). During the 2011 crèche stage no differences in diet were evident with birds consuming similar proportions of rock cod then Patagonian squid (Fig. S4 (B)). During the guard stage of 2012, birds at Cow Bay consumed significantly more rock cod than at Bull Point and Bull Roads where birds consumed a greater proportion of Patagonian squid then lobster Krill (Fig. S4 (C)). Similarly, the large proportion of lobster krill consumed at Bull Point and Bull Roads during the crèche stage of 2012 led to these colonies having similar diet, whereas, birds at Steeple Jason and Cow Bay consumed significantly more Falkland herring which drove the observed difference in diet across colonies for this sampling period (Fig. S4 (D)). In the final guard stage sampled, 2013, an influx of southern blue whiting (*Micromesistius australis*) in the diet of birds at Cow Bay drove a significant difference in diet compared to Bull Roads birds where nearly all prey consumed consisted of lobster krill (Fig. S4 (E)).

Dietary niche metrics

Trophic niche widths were typically lower for birds at Steeple Jason (0.40–0.95) compared to other colonies (1.07–1.51) for all sampling periods except the guard stage of 2012 and 2013 at Cow Bay and Bull Roads, when TNW's were 0.87 and 0.36, respectively. Birds at Steeple Jason also typically demonstrated lower levels in the degree of diet specialisation, *V*, for all periods bar the aforementioned (Table S7). In all sampling periods, except the crèche stage of Cow Bay in 2012, the degree of diet specialisation tended more towards zero showing that individuals tended to use the same resources in the same proportions. Within this generalist pattern, there was a significant and strong, positive relationship ($F_{1,14} = 60.46$, $R^2 = 0.81$, $p < 0.001$) between *V* and TNW (Fig. 2). This indicated that when a large variety of resources were consumed in a given sampling period across all birds so too did each individual bird consume a larger variety of resources.

In contrast, the degree of diet specialization across colonies compared to that within a colony increased after accounting for prey items consumed across the same temporal scale (e.g. all colonies during the guard stage of 2011) (Table 3). This reinforces the difference in diet observed across colonies and the increase was particularly large for the crèche and guard stages of 2012 and 2013 when there were influxes of the pelagic prey items in the diet, Falkland herring and southern blue whiting, respectively.

Inter-colony comparison: stable isotope analysis

In 2011, significant differences were present among colonies for both $\delta^{13}\text{C}$ ($F_{2,59} = 13.39$, $p < 0.001$) and $\delta^{15}\text{N}$

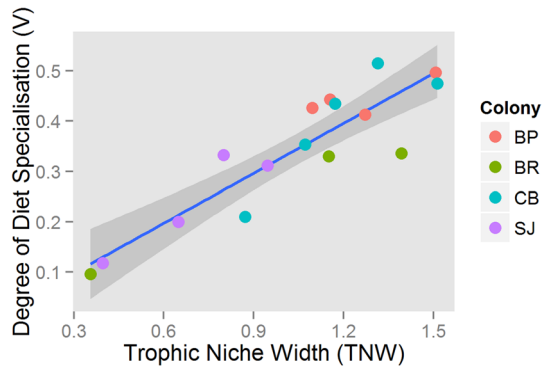


Fig. 2 Linear model indicating relationship between degree of diet specialisation (V) and trophic niche width (TNW) for gentoo penguins sampled from four colonies Bull Point (BP), Bull Roads (BR), Cow Bay (CB) and Steeple Jason (SJ) over three breeding seasons at the Falkland Islands. *Solid line* represents the best fit from linear model with shaded area depicting the 95% confidence bands. Individual data points for the linear model come from sampling season, site and breeding stage specific sampling periods

($F_{2,59} = 26.35, p < 0.001$) (Table 4). Post-hoc testing showed only Steeple Jason had significantly lower values compared to either Cow Bay or Bull Point for $\delta^{13}\text{C}$. For $\delta^{15}\text{N}$, there were significant differences for all pairwise comparisons with the lowest values recorded at Cow Bay followed by Bull Point, then Steeple Jason. These results were illustrated when comparing SEAc, where, Steeple Jason had no overlap with either colony and the SEAc for Cow Bay only encompassed 30% of the larger SEAc for Bull Point (Fig. 3). Regarding ISim and INes, Steeple Jason was clearly separated in isotopic niche space

(ISim/INes = 0/0: SJ vs. CB; = 0.02/0.11: SJ vs. BP), while, Cow Bay was more similar to Bull Roads (ISim/INes = 0.41/0.92: CB vs. BP). The size of the SEA, compared between sites using SEA_B, was significantly larger for Bull Point, then Cow Bay and Steeple Jason (probability < 0.01). Similarly, IRic at Bull Point was twice as large compared to Cow Bay and over five times larger than Steeple Jason (Table 4).

In 2012, significant differences were also present among colonies for both $\delta^{13}\text{C}$ ($F_{3,82} = 100.40, p < 0.001$) and $\delta^{15}\text{N}$ ($F_{3,82} = 15.85, p < 0.001$; Table 4). As in 2011, penguins at Steeple Jason also had significantly lower $\delta^{13}\text{C}$ values in 2012 compared to all colonies. Unlike 2011, only Bull Point and Bull Roads had similar $\delta^{13}\text{C}$ values which were both higher than Cow Bay. Regarding $\delta^{15}\text{N}$, Steeple Jason had significantly higher values than all other colonies, with similar values recorded between Cow Bay and Bull Point and the lowest, but similar values recorded between Bull Point and Bull Roads. In 2012, Steeple Jason had zero overlap with any colonies for both SEAc and ISim (Fig. 3). There was, however, 23% overlap with Cow Bay and the larger SEAc of Bull Point, which overlapped with Bull Roads by 58%. There was no overlap in SEAc between Cow Bay and Bull Roads. Isotopic similarity was strongest between Bull Point and Bull Roads (ISim/INes = 0.49/0.74), then Cow Bay and Bull Point (ISim/INes = 0.25/0.43), and then Cow Bay and Bull Roads (ISim/INes = 0.12/0.24). The size of the SEA_B was most similar between Bull Point, Bull Roads and Cow Bay (all probabilities > 0.4) and least similar for all pairwise comparisons with Steeple Jason (all

Table 3 Degree of individual diet specialisation (V) for breeding gentoo penguins at the Falkland Islands, determined against only the prey items consumed within a single sampling period and against all prey items consumed across the same season (2011–2013) and breeding stage (guard (G) and crèche (C))

Sample period		V		Prey		
Season/stage	Colony	Within	Across	Primary	Secondary	Tertiary
2011 G	SJ	0.31	0.41	PATA	MUN	SAR
	CB	0.35	0.42	PATA	LOL	TG
	BP	0.43	0.49	MUN	PATA	LOL
2011 C	CB	0.44	0.49	PATA	LOL	GON
	BP	0.50	0.51	MUN	PATA	JF
2012 G	CB	0.21	0.34	PATA	LOL	JC
	BP	0.44	0.44	PATA	MUN	LOL
	BR	0.33	0.36	PATA	LOL	MUN
2012 C	SJ	0.33	0.66	SAR	LOL	GON
	CB	0.52	0.62	SAR	PATA	LOL
	BP	0.41	0.61	JF	MUN	PATA
	BR	0.34	0.49	MUN	PATA	JF
2013 G	CB	0.48	0.56	BLU	MUN	LOL
	BR	0.10	0.40	MUN	LOL	PATA

Prey items within each sampling period were identified from the %IRI. See table S5 for prey abbreviations. Note, only those prey with %IRI > 1 were used for the analysis, however, the top three prey items for each period are shown for detail

Table 4 Feather carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) stable isotope values of adult gentoo penguins at the Falkland Islands collected during the breeding seasons of the 2011, 2012 and 2013 austral summers

Year	Colony	n	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	SEAc (‰ ²)	SEAB (‰ ²)	IRic (‰ ²)
2011	BP	20	-15.2 ± 0.5^A	15.7 ± 0.7	0.15	0.14 (0.09/0.22)	0.43
	CB	22	-15.3 ± 0.3^A	15.1 ± 0.6	0.07	0.06 (0.04/0.10)	0.20*
	SJ	20	-15.7 ± 0.2	16.4 ± 0.3	0.03	0.03 (0.02/0.04)	0.08
2012	BP	19	-14.3 ± 0.3^A	$15.6 \pm 0.4^{A,B}$	0.07	0.07 (0.04/0.10)	0.21
	BR	20	-14.1 ± 0.4^A	15.5 ± 0.3^A	0.07	0.06 (0.04/0.10)	0.17*
	CB	28	-14.7 ± 0.3	15.8 ± 0.4^B	0.07	0.07 (0.04/0.09)	0.19*
	SJ	19	-15.7 ± 0.2	16.3 ± 0.5	0.05	0.04 (0.02/0.07)	0.11
2013	BP	20	-14.4 ± 0.3	15.0 ± 0.7^A	0.11	0.09 (0.06/0.20)	0.34
	BR	20	-14.2 ± 0.3	15.4 ± 0.3^A	0.03	0.03 (0.02/0.05)	0.10
	CB	20	-15.1 ± 0.4	15.3 ± 0.4^A	0.07	0.07 (0.05/0.11)	0.16

Collections occurred at Bull Point (BP), Bull Roads (BR), Cow Bay (CB) and Steeple Jason (SJ). Isotopic niche indices include: standard ellipse area corrected for small sample size (SEAc), the Bayesian estimate of the standard ellipse area with lower and upper bounds of 95% credible intervals (SEAB, Jackson et al. 2011) and, isotopic richness (IRic, Cucherousset and Villéger 2015), the area of the convex hull

^{A,B}Periods where samples were non-significantly different

*Average calculated value after bootstrapping to account for varying sample size

probabilities = 0.1) (Fig. 3). A result further reflected by IRic for Steeple Jason being approximately half the value of all other colonies in 2012 (Table 4).

In contrast, the final sampling season, 2013, showed significant differences for $\delta^{13}\text{C}$ ($F_{2,57} = 100.40$, $p < 0.001$) but no significant differences for $\delta^{15}\text{N}$ ($F_{2,57} = 2.62$, $p = 0.080$) among colonies (Table 4). Significance in $\delta^{13}\text{C}$ was strong between Cow Bay and Bull Point ($p < 0.001$) or Bull Roads ($p < 0.001$), however, was marginal between Bull Point and Bull Roads ($p = 0.04$). This is illustrated (Fig. 3) by zero overlap in SEAc for Cow Bay with either colony, and a 16% overlap of the smaller Bull Roads SEAc within that of Bull Point. Cow Bay also had lower overlap in total isotopic niche area compared to Bull Point (ISim/INes = 0.10/0.28) then Bull Roads (ISim/INes = 0.07/0.18), which, had greater overlap between them due to Bull Roads being largely encompassed by Bull Point (ISim/INes = 0.24/0.85). The size of the SEAB (Fig. 3) was similar between Bull Point and Cow Bay (probability = 0.13), however both of these colonies had significantly larger ellipses than Bull Roads (probability < 0.01) (Fig. 3). This was also reflected by Bull Roads having isotopic richness nearly half of Cow Bay and over a third of Bull Point (Table 4).

Inter-colony comparison: isotopic diversity metrics

In all years IDiv approximated 1 (Table 5) indicating that groups were further apart and hence different in isotopic space confirming the lack of overlap for SEAc or the convex hull. Similarly, as IDis also typically tended towards 1 this supported the IDiv results and indicates that across groups there were different isotopic values. Typically, the range of resources used each year across all colonies was higher than for the range of resources used at a specific

colony as indicated by IEve tending towards 1. Notably though, were the typically lower (although all tending towards 1) values of IDiv, IDis and IEve in 2012 which were supported by having the lowest IUni value. This indicates that groups were more similar, which was driven by the similarity amongst sites in niche space overlap, except for Steeple Jason in 2012 (Fig. 3).

Intra-colony comparison: isotopic diversity metrics

Within each colony the typical pattern across all years showed IDis tending towards 0 indicating that individuals within these groups have similar isotopic values (Table 5). This was supported by the IUni values tending towards 0, showing that most of the weight belonged to organisms that are isotopically similar. The typically high IEve values indicate broad resource use over the entire range of the resources within isotopic niche space. The only exception is at Cow Bay in 2013 where IDis tended more towards 1, indicating that individuals within this group had different isotopic values which can also be seen by a lack of individuals within the core isotopic niche area for that year (Fig. 3).

Discussion

Our study is the first detailed multi-year study on gentoo penguin diet at the Falkland Islands. Through a combination of stomach sampling and stable isotope analysis, we found that gentoo penguin diet differed between select breeding colonies around the Islands. These results point toward the important role of available habitat within close proximity to gentoo penguin breeding colonies and the

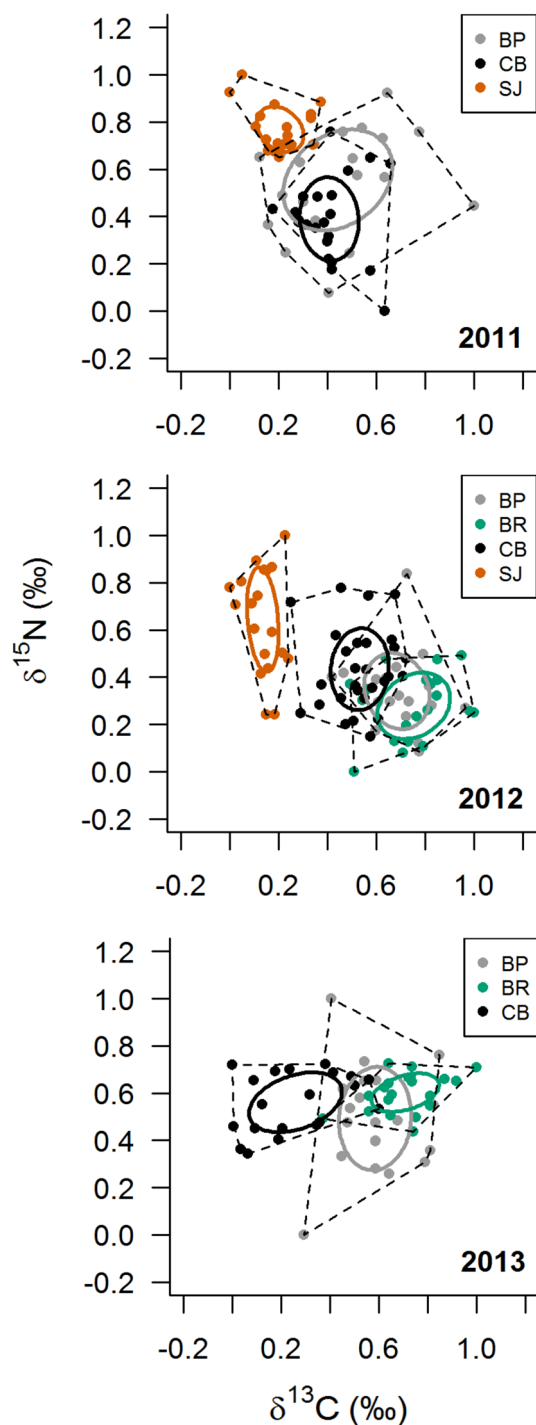


Fig. 3 Feather stable isotope values ($\delta^{13}\text{C}$ and $\delta^{15}\text{N}$, centered and rescaled data) for adult gentoo penguins breeding at four colonies: Bull Point (BP), Bull Roads (BR), Cow Bay (CB) and Steeple Jason (SJ) at the Falkland Islands. Feathers were collected from breeding adult birds during concurrent stomach sampling in the austral summer breeding season: 2011, 2012 and 2013. Total (dashed lines, IRic) and core isotopic niche area adjusted for sample size (solid lines, SEAc) are shown

influence of habitat on prey diversity and consequently gentoo penguin diet.

In summary, six main prey items were of major significance across all sampling periods. These were, in order of importance, rock cod fish, lobster krill, Falkland herring, Patagonian squid, juvenile fish and southern blue whiting. Between different stages within a breeding period, significant difference occurred four out of seven times and when accounting for these different breeding stages, across colony difference in diet occurred four out of five times. Birds from the Steeple Jason colony generally had the narrowest trophic niche width and degree of diet specialization. When looking at all sampling periods, results from the linear model indicate that when gentoo penguins feed on a broader array of prey items so too does each individual bird consume a larger variety of prey items. The differences observed in diet were also reflected in the across colony degree of diet specialization when birds always had higher values than when looking at the within colony component. This was particularly so when birds consumed pelagic prey at some colonies. Clear differences in isotopic values were observed across the colonies with Steeple Jason birds always having the lowest $\delta^{13}\text{C}$ and highest $\delta^{15}\text{N}$ values. These differences were also reflected in the isotopic niche space, where, similarly to TNW, birds from Steeple Jason almost always occupied the smallest, separated niche space, for both SEA_B and IRic. The smallest occupied isotopic niche space recorded was for birds from Bull Roads in 2013 when birds from the Steeple Jason colony were unable to be sampled.

Temporal and spatial diet variation

Prey switching may be necessitated by differing demands for energy requirements during the breeding season (Williams and Rothery 1990; Le corre et al. 2003, Quillfeldt et al. 2011). This is particularly relevant when considering the significant differences in diet that we observed between the guard and crèche stages of the 2012 breeding season. As chicks got older, diet changed from rock cod to either; Falkland herring, a combination of juvenile fish and lobster krill, or lobster krill at Cow Bay, Bull Point and Bull Roads respectively. Of these prey items, Ciancio et al. (2007) showed that rock cod had the lowest energy content by wet mass (4798 j.g^{-1}) compared to Falkland herring (7148 j.g^{-1}) and lobster krill ($11,008 \text{ j.g}^{-1}$, although their measure was for dry mass). Hence, the calorific values of prey are consistent with the hypothesis that gentoo penguins might change diet to meet increased energetic demands of growing chicks, as has been suggested for other penguins (e.g. Jansen et al. 2002; Browne et al. 2011). Notably, when higher energy content prey items are consumed there is often an associated increase in reproductive success, higher

Table 5 Inter and intra-colony isotopic diversity metrics (Cucherousset and Villéger 2015): Isotopic divergence (IDiv), dispersion (IDis), evenness (IEve) and uniqueness (IUni) determined from feathers of breeding adult gentoo penguins at the Falkland Islands

Year	Inter-colony				BP				BR				CB				SJ			
	IDiv	IDis	IEve	IUni	IDiv	IDis	IEve	IUni	IDiv	IDis	IEve	IUni	IDiv	IDis	IEve	IUni	IDiv	IDis	IEve	IUni
2011	0.79	0.71	0.57	0.6	0.76	0.48	0.82	0.32					0.73	0.39	0.71	0.35	0.76	0.42	0.69	0.45
2012	0.63	0.51	0.59	0.47	0.67	0.34	0.76	0.23	0.76	0.49	0.73	0.3	0.72	0.46	0.8	0.3	0.71	0.5	0.76	0.33
2013	0.96	0.94	0.96	0.95	0.71	0.38	0.71	0.28	0.7	0.38	0.76	0.27	0.75	0.61	0.8	0.49				

Samples collected over three years from four different colonies: Bull Point (BP), Bull Roads (BR), Cow Bay (CB) and Steeple Jason (SJ)

fledgling mass and higher growth increments not only for penguins (van Heezik 1990) but also for flying species such as black-legged kittiwakes *Rissa tridactyla* and tufted puffins *Fratercula cirrhata* (Romano et al. 2006).

However, without simultaneous at-sea surveys of prey availability we cannot distinguish whether the prey switching behaviour we report is a result of prey availability, or prey preference. More typically, prey availability, rather than prey preference, is believed to be the driving force behind gentoo penguin diet switching. This is because annual variation in the diet of gentoo penguins (Volkman et al. 1980; Adams and Klages 1989; Coria et al. 2000; Libertelli et al. 2004), and at different sites within an archipelago (Lescroël et al. 2004; Clausen et al. 2005; Miller et al. 2010) has been recorded when there were known differences in prey availability based on at-sea surveys.

In our study, birds at Steeple Jason fed primarily on Falkland herring followed by rock cod, those at Cow Bay fed mainly on rock cod with influxes of Falkland herring and southern blue whiting, and, at the two colonies in the south of the islands, Bull Point and Bull Roads, birds consumed mostly lobster krill then Patagonian squid and rock cod. The locations of these colonies, and their proximity to the shelf edge or position in different water masses appears to play a key role in determining prey availability. Most notable is the major division between the west and east side of the Islands. The Steeple Jason colony, in the west, is situated in a separate water mass, the western branch of the Falkland Current, which is derived from the Antarctic Circumpolar Current (Agnew 2002; Arkhipkin et al. 2010; Ashford et al. 2012). The eastern branch of the Falkland current is stronger, however, both bring upwelled nutrients and create highly productive waters around the islands due to retention of nutrients in localised eddies (Agnew 2002; Arkhipkin et al. 2010; Ashford et al. 2012).

Around the Falklands, Falkland herring move inshore after spawning in September and October (Agnew 2002), with the largest populations being found towards the north-west of the islands during at-sea surveys (Laptikhovskiy et al. 2001; Agnew 2002). This indicates greater availability around Steeple Jason. The other pelagic prey item consumed, southern blue whiting, typically occurred towards

the north-east of the Falkland Islands (Niklitschek et al. 2010; Arkhipkin et al. 2013) during trawls from October to December (Arkhipkin et al. 2013), rendering it a suitable prey item for gentoo penguins at Cow Bay.

Similarly, the presence of the benthic rock cod fish in high proportions for birds at Cow Bay and as a major dietary component for birds in southern colonies is well explained by its distribution. Juvenile individuals (<150 mm) typically utilize the bottom waters over the shelf (Laptikhovskiy and Arkhipkin 2003; Brickle et al. 2006) and recent at-sea surveys found the highest abundance of small fish (<300 mm) towards the north-east in October and north-west in February with fish still in the north-east during this time (Arkhipkin et al. 2013). The juvenile fish also occur in the Patagonian squid fishing grounds (Laptikhovskiy and Arkhipkin 2003) which helps to explain the co-occurrence of these prey items in the diet of gentoo penguins in southern colonies. These Patagonian squid typically concentrate near the bottom during the day, the time when gentoo penguins typically forage, with smaller individuals (dorsal mantle length, DML <80 mm) inhabiting shallow waters, <80 m, and larger individuals (DML 90–100 mm) being found in deeper waters, 100–200 m, which is still over the shelf where the birds typically feed (Masello et al. 2010; Miller et al. 2010). At the Falkland Islands, the squid are abundant to the south and east of the islands, however, the most abundant concentrations, with particular respect to the study colonies, have been found towards the north east (Arkhipkin et al. 2013). Finally, the high consumption of benthic, adult lobster krill in the south of the islands fits well with the surrounding calmer shallow waters as high concentrations of this prey item are better suited to these conditions following a pelagic juvenile dispersal phase (Zeldis 1985; León et al. 2008; Meerhoff et al. 2013).

Our results clearly show that at the scale of the archipelago gentoo penguin diet is diverse. This relates to both the type of prey consumed and the associated habitat utilized by these prey. Hence, it is clear that gentoo penguins can utilise different parts of the water column at various colonies across the Falklands, as documented at other locations (Hindell 1989; Lescroël et al. 2004; Clausen et al. 2005;

Miller et al. 2010). This is understandable as across their range gentoo penguins seldom travel farther than 30 km from the colony during the breeding season (Trivelpiece et al. 1987; Wilson et al. 1998; Miller et al. 2009; Handley 2014). This distance makes large shallow areas of the surrounding shelf readily available for birds at Cow Bay, Bull Point and Bull Roads, but far less accessible to those birds at Steeple Jason where the 100 m isobath is in close proximity to the island. Although gentoo penguins have been recorded to dive as deep as 210 m (Bost et al. 1994), their typical depth range of 30–50 m (Williams et al. 1992; Miller et al. 2009; Kokubun et al. 2010) makes them far more suited to the consumption of benthic prey around the eastern part of the Falkland Islands.

Initial insights from stable isotope values indicate that birds at Steeple Jason may, in particular, be utilizing different areas at sea and foraging at different trophic levels as $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values were always lower and higher, respectively, compared to other colonies. This is in contrast to what one would expect should birds be feeding on similar prey items beyond the breeding period because $\delta^{15}\text{N}$ values are typically higher for rock cod and lobster krill, the benthic prey items (Quillfeldt et al. 2015, Table S2). Furthermore, the conclusion regarding varied habitat use is biased as the $\delta^{13}\text{C}$ isoscape values appear to be higher in the south and east of the Falklands (Quillfeldt et al. 2010), along with the $\delta^{15}\text{N}$ isoscape values typically being lower to the east (Quillfeldt et al. 2015). Additionally, all the $\delta^{13}\text{C}$ values are higher than the average of -16.26‰ for gentoo penguins recorded at New Island on the west of the Falklands (Weiss et al. 2009), nearest to the Steeple Jason study colony. Therefore, similarly to Weiss et al. (2009) we also suggest that gentoo penguins during the pre-moult stage remain foraging in inshore waters. This is further supported by our value for $\delta^{13}\text{C}$ being more similar to that of the imperial cormorant *Phalacrocorax articeps* ($\delta^{13}\text{C} = -15.38\text{‰}$) at New Island, an inshore foraging species (Weiss et al. 2009).

Niche metrics

Polito et al. (2015) provided the first evidence to suggest gentoo penguins, at least within part of their range, are Type B generalists. By incorporating stomach content data representing breeding period diet and insights from stable isotope values of feathers, a proxy for the pre-moult diet, this study supports the proposed idea that gentoo penguins are Type B generalists, particularly when looking at the island wide population.

Deep waters surrounding the Steeple Jason colony probably prevents benthic foraging due to energetic constraints. Therefore, it is unsurprising that bird's at Steeple Jason typically had the lowest degrees of individual

diet specialization, V , and that this degree of specialization increased as the trophic niche width of the birds increased. This increase is likely an artefact of increased availability to multiple foraging opportunities at the other colonies where birds may more feasibly forage benthically or pelagically. The fact that birds at Steeple Jason fed on rock cod, a prey item considered benthic in nature, during the 2011 guard stage, can possibly be explained by the larger rock cod consumed during this time (Table S4). These fish were most likely feeding in pelagic waters as diet of rock cod changes when larger adults consume more planktonic prey compared to juveniles which feed benthically (Laptikhovsky and Arkhipkin 2003).

During the pre-molt period, towards the end of February, both the core isotopic niche space and isotopic richness were also generally lower for birds at Steeple Jason. This may be due to limited dispersal during this time for which no knowledge exists yet as to where birds might be at sea. However, unlike their congeners, gentoo penguins do not migrate large distances during the winter, outside the breeding period (Wilson et al. 1998; Clausen and Pütz 2003; Tanton et al. 2004). Therefore, prior to molting these birds probably remain foraging pelagically in the inshore environment, close to their colony, which would explain their relatively low niche space being occupied. In addition, isotopic divergence and dispersion tended more towards 0 when looking at the intra-colony isotopic diversity metrics showing that within a colony individuals were more similar. This supports the notion that at the colony level across the Falkland Islands gentoo penguins are Type B generalists.

The influence of habitat affecting the realized niche of individuals has been recognized across a wide range of taxa for terrestrial, aquatic and marine organisms such as the gray wolf *Canis lupus* (Darimont et al. 2009), American alligator *Alligator mississippiensis* (Rosenblatt et al. 2015), sea otter *Enhydra lutris* (Newsome et al. 2015), tiger shark *Galeocerdo cuvier* (Simpfendorfer et al. 2001) and bull shark *Carcharhinus leucas* (Matich et al. 2011). It is clear that habitat heterogeneity can place an upper bound on an individual's niche width (Araújo et al. 2011; Schriever and Williams 2013). This habitat heterogeneity, however, is not the only factor that can affect the niche width of a population. Indeed both intra and inter-specific interactions have been recognized to affect the degree of individual diet specialization and thus the niche width of a population (Araújo et al. 2011; Matich et al. 2011). To fully elucidate the role of these factors for gentoo penguins at the Falkland Islands would require simultaneous quantification of prey availability and we strongly advocate for this research in the future.

Conclusions

This study has provided the first, island wide, species specific prey description, for gentoo penguins at the Falkland Islands over three consecutive breeding seasons (2011–2013). The data re-iterate the great flexibility in foraging strategies in gentoo penguins which are largely driven by local conditions. This probably to a large extent explains the resilience to environmental change observed in this species (with an increasing global population as opposed to its congeners which are dietary specialists across their range; see Borboroglu and Boersma (2012)). Furthermore, the study highlights how gentoo penguins would require a multi-faceted approach towards their conservation and management owing to spatial variability in prey consumption. Finally, in our attempts to identify prey consumed during the pre-molt period through the use of stable isotope mixing models we were unsuccessful. Critically, future work should focus on determining more prey specific trophic enrichment factors and if possible try to source prey items from a more inshore environment to facilitate the use of stable isotope mixing models towards diet reconstruction.

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Compliance with ethical standards

Conflict of interest The authors declare that there is no conflict of interest and that consent was obtained from all parties. The research was conducted under Grants from the Falkland Islands Environmental Planning Department: R17/2011 & R13/2012.

Human and animal ethics Animal ethics approval was granted by Nelson Mandela Metropolitan University ethics committee (ALL-SCI-ZOO-014).

References

- Adams NJ, Klages NT (1989) Temporal variation in the diet of the gentoo penguin *Pygoscelis papua* at sub-Antarctic Marion Island. *Colon Waterbirds* 12:30–36
- Agnew DJ (2002) Critical aspects of the Falkland Islands pelagic ecosystem—distribution, spawning and migration of pelagic animals in relation to oil exploration. *Aquatic Conserv Mar Freshw Ecosyst* 50:39–50
- Araújo MS, Bolnick DI, Layman CA (2011) The ecological causes of individual specialisation. *Ecol Lett* 14:948–958
- Arkhipkin A, Brickle P, Laptikhovskiy V (2010) The use of island water dynamics by spawning red cod, *Salilota australis* (Pisces: Moridae) on the Patagonian Shelf (Southwest Atlantic). *Fish Res* 105:156–162
- Arkhipkin A, Brickle P, Vladimir L (2013) Links between marine fauna and oceanic fronts on the Patagonian Shelf and Slope. *Arquipel Life Mar Sci* 30:19–37
- Ashford JR, Fach BA, Arkhipkin AI, Jones CM (2012) Testing early life connectivity supplying a marine fishery around the Falkland Islands. *Fish Res* 121:144–152
- Augé AA, Lascelles B, Dias M (2015). Marine Spatial Planning for the Falkland Islands. In: Methodology for identification of important areas for marine megafauna workshop report. South Atlantic Environmental Research Institute, Stanley, Falkland Islands
- Barrett RT, Camphuysen KCJ, Anker-Nilssen T, Chardine JW, Furness RW, Garthe S, Huppoo O, Leopold MF, Montevecchi WA, Veit RR (2007) Diet studies of seabirds: A review and recommendations. *ICES J Mar Sci* 64:1675–1691
- Baylis A, Crofts S, Wolfaardt AC (2013) Population trends of Gentoo penguins *Pygoscelis papua* breeding at the Falkland Islands. *Mar Ornithol* 41:1–5
- Bearhop S, Adams CE, Waldron S, Fuller R, Macleod H (2004) Determining trophic niche width: A novel approach using stable isotope analysis. *J Anim Ecol* 73:1007–1012
- Bolnick DI, Svanbäck R, Fordyce J, Yang LH, Davis JM, Hulsey CD, Forister ML (2003) The ecology of individuals: incidence and implications of individual specialization. *Am Nat* 161:1–28
- Bond AL, Jones IL (2009) A practical introduction to stable-isotope analysis for seabird biologists: approaches, cautions and caveats. *Mar Ornithol* 37:183–188
- Borboroglu PG, Boersma PD (2012) *Penguins: Natural History and Conservation*. University of Washington Press, Seattle
- Bost CA, Lage J, Pütz K (1994) Maximum diving depth and diving patterns of the Gentoo penguin *Pygoscelis papua* at the Crozet Islands. *Mar Ornithol* 22:237–244
- Brickle P, Laptikhovskiy V, Arkhipkin A, Portela J (2006) Reproductive biology of *Patagonotothen ramsayi* (Regan, 1913) (Pisces: Nototheniidae) around the Falkland Islands. *Polar Biol* 29:570–580
- Brickle P, Arkhipkin AI, Laptikhovskiy V, Stocks A, Taylor A (2009) Resource partitioning by two large planktivorous fishes *Micromesistius australis* and *Macruronus magellanicus* in the Southwest Atlantic. *Estuar Coast Shelf Sci* 84:91–98
- Browne T, Lalas C, Mattern T, Van heezik Y (2011) Chick starvation in yellow-eyed penguins: Evidence for poor diet quality and selective provisioning of chicks from conventional diet analysis and stable isotopes. *Austral Ecol* 36:99–108
- Ceia FR, Ramos JA (2015) Individual specialization in the foraging and feeding strategies of seabirds: a review. *Mar Biol* 162:1923–1938
- Ceia FR, Phillips RA, Ramos JA, Chérel Y, Vieira RP, Richard P, Xavier JC (2012) Short- and long-term consistency in the foraging niche of wandering albatrosses. *Mar Biol* 159:1581–1591
- Chao A (1987) Estimating the population size for capture-recapture data with unequal catchability. *Biometrics* 43:783–791
- Chérel Y, Hobson K (2007) Geographical variation in carbon stable isotope signatures of marine predators: a tool to investigate their foraging areas in the Southern Ocean. *Mar Ecol Prog Ser* 329:281–287

- Cherel Y, Hobson K, Weimerskirch H (2000) Using stable-isotope analysis of feathers to distinguish moulting and breeding origins of seabirds. *Oecologia* 122:155–162
- Chouvelon T, Spitz J, Cherel Y, Caurant F, Sirmel R, Mendez-Fernandez P, Bustamante P (2011) Species and ontogenic-related differences in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ values and Hg and Cd concentrations of cephalopods. *Mar Ecol Prog Ser* 433:107–120
- Ciancio JE, Pascual M, Beauchamp D (2007) Energy density of Patagonian aquatic organisms and empirical predictions based on water content. *Trans Am Fish Soc* 136:1415–1422
- Clarke MR (1986) A Handbook for the identification of cephalopod beaks. Clarendon Press, University California, USA
- Clausen AP, Pütz K (2002) Recent trends in diet composition and productivity of gentoo, Magellanic and rockhopper penguins in the Falkland Islands. *Aquat Conserv Mar Freshw Ecosyst* 12:51–61
- Clausen A, Pütz K (2003) Winter diet and foraging range of gentoo penguins (*Pygoscelis papua*) from Kidney Cove, Falkland Islands. *Polar Biol* 26:32–40
- Clausen AP, Arkhipkin AI, Laptikhovskiy V, Huin N (2005) What is out there: diversity in feeding of gentoo penguins (*Pygoscelis papua*) around the Falkland Islands (Southwest Atlantic). *Polar Biol* 28:653–662
- Connan M, McQuaid CD, Bonnevie BT, Smale MJ, Cherel Y (2014) Combined stomach content, lipid and stable isotope analyses reveal spatial and trophic partitioning among three sympatric albatrosses from the Southern Ocean. *Mar Ecol Prog Ser* 497:259–272
- Connan M, Hofmeyr GG, Pistorius PA (2016) Reappraisal of the Trophic Ecology of One of the World's Most Threatened Spheniscids, the African Penguin. *PLoS one* 11(7):e0159402
- Coria N, Libertelli M, Casaux R, Darrieu C (2000) Inter-annual variation in the autumn diet of the gentoo penguin at Laurie Island, Antarctica 23:511–517 (Waterbirds)
- Corman AM, Mendel B, Voigt CC, Garthe S (2016) Varying foraging patterns in response to competition? A multicolony approach in a generalist seabird. *Ecol Evol* 4:974–976
- Cortes E (1997) A critical review of methods of studying fish feeding based on analysis of stomach contents: application to elasmobranch fishes. *Can J Fish Aquat Sci* 54:726–738
- Cucherousset J, Villéger S (2015) Quantifying the multiple facets of isotopic diversity: New metrics for stable isotope ecology. *Ecol Indic* 56:152–160
- Darimont CT, Paquet PC, Remchen TE (2009) Landscape heterogeneity and marine subsidy generate extensive intrapopulation niche diversity in a large terrestrial vertebrate. *J Anim Ecol* 78:126–133
- Duffy DC, Jackson S (1986) Diet studies of seabirds: a review of methods. *Colon Waterbirds* 9:1–17
- Granadeiro JP, Silva MA (2000). The use of otoliths and vertebrae in the identification and size-estimation of fish in predator-prey studies. *Cybius* 24: 383–393
- Handley (2014) Foraging ecology of the gentoo penguin at the Falkland Islands. MSc dissertation, Nelson Mandela Metropolitan University, South Africa
- Handley JM, Baylis AMM, Brickle P, Pistorius P (2015) Temporal variation in the diet of gentoo penguins at the Falkland Islands. *Polar Biol* 39:283–296
- Hindell M (1989) The Diet of Gentoo Penguins *Pygoscelis papua* at Macquarie Island: Winter and Early Breeding Season. *Emu* 2:71–78
- Hobson K, Piatt JF, Pitocchelli J (1994) Using stable isotopes to determine seabird trophic relationships. *J Anim Ecol* 63:786–798
- Huin N (2005) Falkland Islands Seabird Monitoring Programme Annual Report 2003/2004/2005. Falkland Islands
- Inger R, Bearhop S (2008) Applications of stable isotope analyses to avian ecology. *Ibis* 150:447–461
- Jackson AL, Inger R, Parnell AC, Bearhop S (2011) Comparing isotopic niche widths among and within communities: SIBER—Stable Isotope Bayesian Ellipses in R. *J Anim Ecol* 80:595–602
- Jaeger A, Connan M, Richard P, Cherel Y (2010) Use of stable isotopes to quantify seasonal changes of trophic niche and levels of population and individual specialisation in seabirds. *Mar Ecol Prog Ser* 401:269–277
- Jansen J K, Russell RW, Meyer WR (2002) Seasonal shifts in the provisioning behavior of chinstrap penguins, *Pygoscelis antarctica*. *Oecologia* 131 : 306–318
- Jobling M, Breiby A (1986) The use and abuse of fish otoliths in studies of feeding habits of marine piscivores. *Sarsia* 71:265–274
- Karnovsky N, Hobson K, Iverson S (2012) From lavage to lipids: estimating diets of seabirds. *Mar Ecol Prog Ser* 451:263–284
- Kiszka JJ, Aubail A, Hussey NE, Heithaus MR, Caurant F, Bustamante P (2015) Plasticity of trophic interactions among sharks from the oceanic south-western Indian Ocean revealed by stable isotope and mercury analyses. *Deep Sea Res Part I Oceanogr Res Pap* 96:49–58
- Kokubun N, Takahashi A, Mori Y, Watanabe S, Shin HC (2010) Comparison of diving behavior and foraging habitat use between chinstrap and gentoo penguins breeding in the South Shetland Islands, Antarctica. *Mar Biol* 157:811–825
- Laptikhovskiy V, Arkhipkin A (2003) An impact of seasonal squid migrations and fishing on the feeding spectra of subantarctic notothenioids *Patagonotothen ramsayi* and *Cottoperca gobio* around the Falkland Islands. *J Appl Ichthyol* 19:35–39
- Laptikhovskiy V, Arkhipkin A, Henderson C (2001) Feeding habits and dietary overlap in spiny dogfish *Squalus acanthias* (Squalidae) and narrowmouth catshark *Schroederidhthys bivius* (Scyliorhinidae). *J Mar Biol Assoc U K* 81:1015–1018
- Le corre M, Cherel Y, Lagarde F, Lormée H, Jouventin P (2003) Seasonal and inter-annual variation in the feeding ecology of a tropical oceanic seabird, the red-tailed tropicbird *Phaethon rubricauda*. *Mar Ecol Prog Ser* 255:289–301
- León R, Castro LR, Cáceres M (2008) Dispersal of *Munida gregaria* (Decapoda: Galatheidae) larvae in Patagonian channels of southern Chile. *ICES J Mar Sci* 65:1131–1143
- Lescroëil A, Bost CA (2005) Foraging under contrasting oceanographic conditions: the gentoo penguin at Kerguelen Archipelago. *Mar Ecol Prog Ser* 302:245–261
- Lescroëil A, Ridoux V, Bost CA (2004) Spatial and temporal variation in the diet of the gentoo penguin (*Pygoscelis papua*) at Kerguelen Islands. *Polar Biol* 27:206–216
- Libertelli M, Daneri GA, Piatkowski U, Coria NR, Carlini AR (2004) Predation on cephalopods by *Pygoscelis papua* and *Arctocephalus gazella* at South Orkney Islands. *Pol. Polar Res* 25:267–274
- Lynch HJ (2012) The Gentoo penguin (*Pygoscelis papua*). In: Borborough PG, Boersma PD (eds.) *Biology and Conservation of the World's penguins*. University of Washington, Seattle
- Masello JF, Mundry R, Poisbleau M, Demongin L, Voigt CC, Wikelski M, Quillfeldt P (2010) Diving seabirds share foraging space and time within and among species. *Ecosphere* 1:1–28
- Match P, Heithaus MR, Layman C (2011) Contrasting patterns of individual specialization and trophic coupling in two marine apex predators. *J Anim Ecol* 80:294–305
- Meerhoff E, Castro L, Tapia F (2013) Influence of freshwater discharges and tides on the abundance and distribution of larval and juvenile *Munida gregaria* in the baker river estuary, Chilean patagonia. *Cont Shelf Res* 61–62:1–11
- Miller AK, Karnovsky NJ, Trivelpiece WZ (2009) Flexible foraging strategies of gentoo penguins *Pygoscelis papua* over 5 years in the South Shetland Islands, Antarctica. *Mar Biol* 156:2527–2537
- Miller AK, Kappes M, Trivelpiece SG, Trivelpiece WZ (2010) Foraging-Niche Separation of Breeding Gentoo and

- Chinstrap Penguins, South Shetland Islands, Antarctica. *Condor* 112:683–695
- Mizutani H, Fukuda M, Kabaya Y, Wada E (1990) Carbon isotope ratio of feathers reveals feeding behavior of cormorants. *Auk* 107:400–403
- Newsome SD, Tinker MT, Gill V, Hoyt ZN, Doroff A, Nichol L, Bodkin JL (2015) The interaction of intraspecific competition and habitat on individual diet specialization: a near range-wide examination of sea otters. *Oecologia* 178:45–59
- Niklitschek EJ, Secor DH, Toledo P, Lafon A, George-Nascimento M (2010) Segregation of SE Pacific and SW Atlantic southern blue whiting stocks: Integrating evidence from complementary otolith microchemistry and parasite assemblage approaches. *Environ Biol Fishes* 89:399–413
- Parnell AC, Inger R, Bearhop S, Jackson AL (2010) Source partitioning using stable isotopes: Coping with too much variation. *PLoS One* 5:1–5
- Phillips DL, Newsome SD, Gregg JW (2005) Combining sources in stable isotope mixing models: alternative methods. *Oecologia* 144:520–527
- Pinkas LM, Oliphant MS, Iverson ILK (1971) Food habits of albacore, Bluefin tuna and bonito in California waters. *Fish Bull Calif Dep Fish Game* 152:1–105
- Polito MJ, Abel S, Tobias CR, Emslie SD (2011a) Dietary isotopic discrimination in gentoo penguin (*Pygoscelis papua*) feathers. *Polar Biol* 34:1057–1063
- Polito MJ, Trivelpiece WZ, Karnovsky NJ, Ng E, Patterson WP, Emslie SD (2011b) Integrating stomach content and stable isotope analyses to quantify the diets of pygoscelid penguins. *PLoS One* 6:e26642
- Polito M, Trivelpiece W, Patterson W, Karnovsky N, Reiss C, Emslie S (2015) Contrasting specialist and generalist patterns facilitate foraging niche partitioning in sympatric populations of Pygoscelis penguins. *Mar Ecol Prog Ser* 519:221–237
- Post DM, Layman C, Arrington DA, Takimoto G, Quattrochi J, Montaña CG (2007) Getting to the fat of the matter: models, methods and assumptions for dealing with lipids in stable isotope analyses. *Oecologia* 152:179–189
- Pütz K, Ingham RJ, Smith JG, Croxall JP (2001) Population trends, breeding success and diet composition of gentoo *Pygoscelis papua*, magellanic *Spheniscus magellanicus* and rockhopper *Eudyptes chrysocome* penguins in the Falkland Islands. *Polar Biol* 24:793–807
- Quillfeldt P, Masello JF, McGill RA, Adams M, Furness RW (2010) Moving polewards in winter: a recent change in the migratory strategy of a pelagic seabird? *Front Zool* 7:15
- Quillfeldt P, Masello JF, Brickle P, Martin-cruzburg D (2011). Fatty acid signatures of stomach contents reflect inter-and intra-annual changes in diet of a small pelagic seabird, the Thin-billed prion *Pachyptila belcheri*. *Mar Biol* 158: 1805–1813
- Quillfeldt P, Ekschmitt K, Brickle P, McGill RR, Wolters V, Dehnhard N, Masello JF (2015) Variability of higher trophic level stable isotope data in space and time - a case study in a marine ecosystem. *Rapid Commun Mass Spectrom* 29:667–674
- R Development Core Team (2015) R: a language and environment for statistical computing. (2015) R Foundation for Statistical Computing, Vienna, <http://www.r-project.org>
- Ratcliffe N, Trathan PN (2011) A review of the diet and at-sea distribution of penguins breeding within the CCAMLR Convention Area. *CCAMLR Sci* 19:75–114
- Romano MD, Piatt JF, Roby DD (2006) Testing the junk-food hypothesis on marine birds: effects of prey type on growth and development. *Waterbirds* 29:407–414
- Rosenblatt AE, Nifong JC, Heithaus MR, Mazzotti FJ, Cherkiss MS, Jeffery BM, Elsey RM, Decker R, Silliman BR, Guillette LJ, Lowers RH, Larson JC (2015) Factors affecting individual foraging specialization and temporal diet stability across the range of a large “generalist” apex predator. *Oecologia* 178:5–16
- Rosenzweig ML (2007) On foraging theory, humans, and the conservation of diversity. In: Stephens DW, Brown JS, Ydenberg RC (eds.) *Foraging: Behaviour and Ecology*. University of Chicago, Chicago, p 608
- Roughgarden J (1979) *Theory of population genetics and evolutionary ecology: an introduction*. Macmillan, New York
- Schriever TA, Williams D (2013) Ontogenetic and individual diet variation in amphibian larvae across an environmental gradient. *Freshw Biol* 58:223–236
- Simpfendorfer CA, Goodreid AB, Mcauley RB (2001) Size and geographic variation in the diet of the tiger shark, *Galeocerdo cuvier*, from Western Australian waters. *Environ Biol Fishes* 61:37–46
- Smith J, Mazumder D, Suthers IM, Taylor MD (2013) To fit or not to fit: Evaluating stable isotope mixing models using simulated mixing polygons. *Methods Ecol Evol* 4:612–618
- Stanworth A (2013) Falkland Islands Seabird Monitoring Programme Annual Report 2012/13. Falkland Islands
- Tanton JL, Reid K, Croxall JP, Trathan PN (2004) Winter distribution and behaviour of gentoo penguins *Pygoscelis papua* at South Georgia. *Polar Biol* 27:299–303
- Tapella F, Lovrich GA (2006) Morphological differences between “subrugosa” and “gregaria” morphs of adult *Munida* (Decapoda): *Anomura*: Galatheididae) from the Beagle Channel, southern South America. *J Mar Biol Assoc United Kingdom* 86:1149–1155
- Thomson J, Heithaus MR, Burkholder D, Vaudo JJ, Wirsing AJ, Dill LM (2012) Site specialists, diet generalists? Isotopic variation, site fidelity, and foraging by loggerhead turtles in Shark Bay, Western Australia. *Mar Ecol Prog Ser* 453:213–226
- Trivelpiece WZ, Bengtson JL, Trivelpiece SG, Volkman NJ (1986) Foraging behavior of Gentoo and Chinstrap penguins as determined by new radiotelemetry techniques. *Auk* 103:777–781
- Trivelpiece WZ, Trivelpiece SG, Volkman NJ (1987) Ecological segregation of Adélie, gentoo, and chinstrap penguins at King George Island, Antarctica. *Ecology* 68:351–361
- Van Valen L (1965) Morphological Variation and Width of Ecological Niche. *Am Nat* 99:377–390
- Van Heezik Y (1990) Seasonal, geographical, and age-related variations in the diet of the yellow-eyed penguin (*Megadyptes antipodes*). *New Zeal. J Zool* 17:201–212
- Van Heezik Y, Seddon PJ (1989) Stomach sampling in the Yellow-eyed penguin: Erosion of otoliths and squid beaks (Erosión de otolitos y picos de calamares en el estómago de Pingüinos (*Megadyptes antipodes*)). *J F Ornithol* 60:451–458
- Volkman NJ, Presler P, Trivelpiece W (1980) Diets of Pygoscelid Penguins at King George Island, Antarctica. *Condor* 82:373–378
- Weiss F, Furness RW, McGill R, Strange IJ, Masello JF, Quillfeldt P (2009) Trophic segregation of Falkland Islands seabirds: insights from stable isotope analysis. *Polar Biol* 32:1753–1763
- Williams TD, Rothery P (1990) Factors affecting variation in foraging and activity patterns of gentoo penguins (*Pygoscelis papua*) during the breeding season at Bird Island, South Georgia. *J Appl Ecol* 27:1042–1054
- Williams TD, Kato A, Croxall JP, Naito Y, Briggs DR, Barton TR, Rodwell S, Barton TIMR (1992) Diving Pattern and Performance in Nonbreeding Gentoo Penguins (*Pygoscelis papua*) during Winter. *Auk* 109:223–234
- Wilson RP (1984) An improved stomach pump for penquins and other seabirds. *J F Ornithol* 55:109–112
- Wilson RP, Alvarez B, Latorre L, Adelung D, Culik B, Bannasch R (1998) The movements of gentoo penguins *Pygoscelis papua* from Ardley Island, Antarctica. *Polar Biol* 19:407–413

- Xavier JC, Cherel Y (2009) Cephalopod Beak Guide for the Southern Ocean. British Antarctic Survey, Cambridge
- Zaccarelli N, Bolnick DI, Mancinelli G (2013) RInSp: a package for the analysis of individual specialization in resource use. *Methods Ecol Evol* 4:1018–1023
- Zeldis JR (1985) Ecology of *Munida gregaria* (Decapoda, Anomura): distribution and abundance, population dynamics and fisheries. *Oldendorf* 22:77–99
- Zenteno L, Borella F, Otero JG, Piana E, Belardi JB, Borrero LA, Saporiti F, Cardona L, Crespo E (2015) Shifting niches of marine predators due to human exploitation: the diet of the South American sea lion (*Otaria flavescens*) since the late Holocene as a case study. *Paleobiology* 41:387–401
- Zuur AF, Ieno EN, Walker NJ, Saveliev AA, Smith GM (2009) Mixed effects models and extensions in ecology with R. Springer Science and Business Media, New York