

# Stable isotopes demonstrate intraspecific variation in habitat use and trophic level of non-breeding albatrosses

WILLIAM F. MILLS,<sup>1,2\*</sup>  RONA A. R. MCGILL,<sup>3</sup>  YVES CHEREL,<sup>4</sup>  STEPHEN C. VOTIER<sup>2</sup>  & RICHARD A. PHILLIPS<sup>1</sup>

<sup>1</sup>British Antarctic Survey, Natural Environment Research Council, Cambridge, CB3 0ET, UK

<sup>2</sup>Centre for Ecology and Conservation, Penryn Campus, University of Exeter, Penryn, Cornwall, TR10 9EZ, UK

<sup>3</sup>NERC Life Sciences Mass Spectrometry Facility, Scottish Universities Environmental Research Centre, East Kilbride, G75 0QF, UK

<sup>4</sup>Centre d'Etudes Biologiques de Chizé (CEBC), UMR 7372 du CNRS- La Rochelle Université, Villiers-en-Bois, 79360, France

The non-breeding period is critical for restoration of body condition and self-maintenance in albatrosses, yet detailed information on diet and distribution during this stage of the annual cycle is lacking for many species. Here, we use stable isotope values of body feathers ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) to infer habitat use and trophic level of non-breeding adult Grey-headed Albatrosses *Thalassarche chrystostoma* ( $n = 194$ ) from South Georgia. Specifically, we: (1) investigate intrinsic drivers (sex, age, previous breeding outcome) of variation in habitat use and trophic level; (2) quantify variation among feathers of the same birds; and (3) examine potential carry-over effects of habitat use and trophic level during the non-breeding period on subsequent breeding outcome. In agreement with previous tracking studies,  $\delta^{13}\text{C}$  values of individual feathers indicate that non-breeding Grey-headed Albatrosses from South Georgia foraged across a range of oceanic habitats, but mostly in subantarctic waters, between the Antarctic Polar Front and Subtropical Front. Sex differences were subtle but statistically significant, and overlap in the core isotopic niche areas was high (62%); however, males exhibited slightly lower  $\delta^{13}\text{C}$  and higher  $\delta^{15}\text{N}$  values than females, indicating that males forage at higher latitudes and at a higher trophic level. Neither age nor previous breeding outcome influenced stable isotope values, and we found no evidence of carry-over effects of non-breeding habitat use or trophic level on subsequent breeding outcome. Repeatability among feathers of the same individual was moderate in  $\delta^{13}\text{C}$  and low in  $\delta^{15}\text{N}$ . This cross-sectional study demonstrates high variability in the foraging and migration strategies of this albatross population.

**Keywords:** intraspecific variation, migration, Procellariiformes, seabirds, Southern Ocean, *Thalassarche chrystostoma*.

Individual seabirds within apparently generalist populations may differ in their resource use and foraging strategies (Phillips *et al.* 2017), with potential implications for community structure, evolutionary ecology and conservation

management (Bolnick *et al.* 2003, Votier *et al.* 2010, Phillips *et al.* 2017). Variation among individuals is often attributed to the influence of intrinsic drivers, including sex (Bearhop *et al.* 2006, Phillips *et al.* 2011), age (Votier *et al.* 2017, Clay *et al.* 2018) and breeding status (Jaeger *et al.* 2014, Clay *et al.* 2016). However, most studies of intraspecific variation in seabird foraging have focused on the breeding period and less is known

\*Corresponding author.  
Email: wilmil23@bas.ac.uk  
Twitter: @WF\_Mills

about the non-breeding period, when birds are free from reproductive constraints associated with incubation or provisioning, and can disperse widely.

There is increasing evidence for sex differences in seabird foraging strategies during the non-breeding period, including variation in diet, distribution and other aspects of behaviour (Bearhop *et al.* 2006, Phillips *et al.* 2009, De Felipe *et al.* 2019). These differences are thought to arise from size-related behavioural dominance and competitive exclusion (typically of the larger sex over the smaller) or habitat and dietary specialization (Phillips *et al.* 2017). The degree of segregation in foraging ecology may be a function of sexual size dimorphism (SSD; Phillips *et al.* 2011), although this appears to apply more to high-latitude than to tropical species (Mancini *et al.* 2013), and studies have also demonstrated sex-related foraging strategies in species with reduced SSD (e.g. Lamacchia *et al.* 2019). Less attention, however, has been paid to changes in foraging behaviour and distribution with age, despite some evidence that it may be an important driver (Phillips *et al.* 2017). Indeed, a recent study of non-breeding Wandering Albatrosses *Diomedea exulans* from South Georgia found evidence of age-related changes in activity patterns and stable isotope proxies for habitat use (Clay *et al.* 2018). There is also evidence that non-breeding Wandering Albatrosses in the Indian Ocean forage further south with age (Jaeger *et al.* 2014).

Stable isotope analyses have been validated as a powerful tool to study the foraging ecology of seabirds, including albatrosses (Cherel *et al.* 2000), and stable isotope values of adult feathers are routinely used to investigate diet and distribution during the non-breeding period (Phillips *et al.* 2009, Cherel *et al.* 2013). This is because moult rarely overlaps with breeding, and feathers are metabolically inert once grown and hence preserve an isotopic record of diet at the time of formation (Hobson & Clark 1992, Bearhop *et al.* 2002, Catry *et al.* 2013). Stable isotope values of carbon ( $^{13}\text{C}/^{12}\text{C}$ , expressed as  $\delta^{13}\text{C}$ ) and nitrogen ( $^{15}\text{N}/^{14}\text{N}$ , expressed as  $\delta^{15}\text{N}$ ) in tissues reflect those of prey during the period of synthesis.  $\delta^{15}\text{N}$  values increase in a stepwise manner by  $\sim 3\text{--}5\text{‰}$  at each trophic level, whereas  $\delta^{13}\text{C}$  values increase less with trophic level ( $\sim 0.5\text{--}1\text{‰}$ ) but can be used to infer foraging habitat (DeNiro & Epstein 1981, Hobson & Clark 1992, Bearhop *et al.* 2002). In the marine environment, this includes the relative

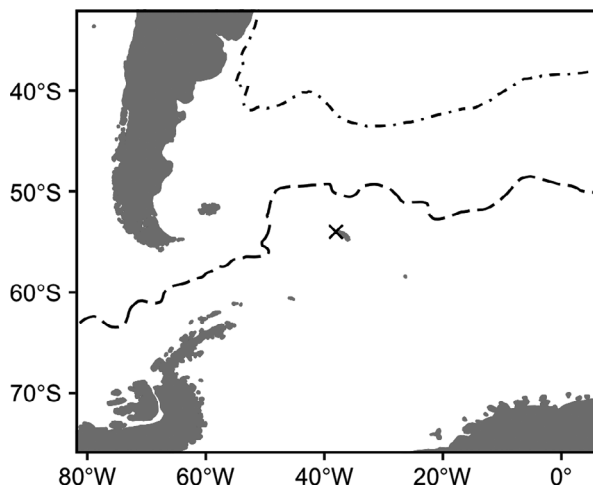
reliance on an inshore vs. offshore, benthic vs. pelagic diet, and latitude or water mass where a gradient exists, such as in the Southern Ocean (Quillfeldt *et al.* 2005, 2010, Cherel & Hobson 2007, Jaeger *et al.* 2010).

The aim of this cross-sectional study is to provide detailed information on the foraging ecology of Grey-headed Albatrosses *Thalassarche chrysotoma* from South Georgia during the non-breeding period. The population at South Georgia constitutes the largest population at any island group (50% globally; Poncet *et al.* 2017). This species exhibits male-biased SSD (Phillips *et al.* 2004), is among the longest-lived birds (Catry *et al.* 2006) and is a population-level generalist (Mills *et al.* 2020). Two studies analysing geolocator data from a small number of non-breeding birds revealed that most target the Sub-Antarctic Zone (SAZ: between the Antarctic Polar Front (APF) and the Subtropical Front (STF)), with evidence for small-scale sexual segregation and some consistency in habitat use within the same non-breeding period (Croxall *et al.* 2005, Clay *et al.* 2016). Using the stable isotope method, we test the following a priori predictions: (1) most feather  $\delta^{13}\text{C}$  values should correspond to foraging in the SAZ; (2) there will be sex differences in stable isotope values, consistent with the degree of SSD; (3)  $\delta^{13}\text{C}$  values will decrease with age, as demonstrated in similar studies of Wandering Albatrosses (Jaeger *et al.* 2014, Clay *et al.* 2018); (4) stable isotope values of feathers sampled from the same individual should show high variability given the protracted moult pattern in this species (see below); and (5) there will be carry-over effects of variation in diet and distribution. Previous breeding outcome may influence the stable isotope values, or the latter may influence the subsequent breeding outcome; however, without further information we make no a priori predictions about the directions of these relationships.

## METHODS

### Study site and colony

Chicks have been ringed annually since the 1970s in the intensive study-colonies of Grey-headed Albatrosses at Bird Island, South Georgia (54°00'S, 38°03'W) (Fig. 1). Daily to weekly visits were made throughout the breeding season to record identities of breeders and non-breeders, laying and



**Figure 1.** Location of Bird Island, South Georgia (black cross), in relation to the Antarctic Polar Front (dashed line) and Subtropical Front (dot-dash line).

fledging dates, and nest survival (Froy *et al.* 2017, Pardo *et al.* 2017), and the modal age of first breeding at Bird Island is 12 years (Prince *et al.* 1994). All birds included in this study were sexed from records of observed copulatory position, from pre-laying attendance pattern or using DNA extracted from blood samples (Fridolfsson & Ellegren 1999); hence all birds were of known age, sex and breeding history.

### Feather sampling and moult

A random collection of relatively unworn body feathers were obtained from the breast region of adults during the early chick-rearing period (December–January) in three separate breeding seasons (2013/14–2015/16). Individuals ( $n = 194$ ) were only sampled in a single season. Stable isotope values of feathers can provide information on foraging ecology during the non-breeding period with the underlying assumption that moult and breeding do not overlap. In a study of albatross moulting patterns at South Georgia, no Grey-headed Albatrosses were in active body feather moult at Bird Island between October and February (Prince *et al.* 1993). However, albatrosses replace their body feathers gradually throughout the non-breeding period; indeed, only ~7% of body feathers may be moulted and regrown at any one time (Battam *et al.* 2010). Therefore, the exact timing of body feather synthesis during the non-breeding period (of ~16 months) is unknown.

However, as our sampling was of relatively unworn feathers, most were likely to have grown in the immediately preceding winter.

### Stable isotope analysis

Three body feathers were selected per individual and analysed separately. Whole feathers were analysed, excluding only the rachis. Feathers were cleaned of surface lipids and contaminants using a chloroform/methanol solution (2 : 1 v/v) followed by successive distilled water rinses. Feathers were air-dried, cut into small fragments using stainless steel scissors and packed into tin capsules (aliquots:  $0.70 \pm 0.01$  mg (mean  $\pm$  standard error)). Stable isotope analyses were conducted at the Natural Environment Research Council (NERC) Life Sciences Mass Spectrometry Facility in East Kilbride. Stable isotope ratios of carbon and nitrogen were determined by a continuous-flow mass spectrometer (Delta Plus XP; Thermo Scientific, Bremen, Germany) coupled to an elemental analyser (vario PYRO cube; Elementar, Langensfeld, Germany). To correct for instrument drift, three internal laboratory standards were analysed for every 10 samples. Stable isotope ratios are reported as  $\delta$ -values and expressed as ‰ according to the equation:  $\delta X = [(R_{\text{sample}}/R_{\text{standard}}) - 1] \times 10^3$ , where X is  $^{13}\text{C}$  or  $^{15}\text{N}$ , R is the corresponding ratio  $^{13}\text{C}/^{12}\text{C}$  or  $^{15}\text{N}/^{14}\text{N}$ , and  $R_{\text{standard}}$  is the ratio of international references Vienna PeeDee Belemnite for carbon and atmospheric  $\text{N}_2$  (AIR) for nitrogen. Measurement precision (standard deviation associated with replicate runs of USGS40) was  $\pm 0.1\text{‰}$  for  $\delta^{13}\text{C}$  and  $\pm 0.2\text{‰}$  for  $\delta^{15}\text{N}$ .

### Data analysis

Stable isotope values were compared with  $\delta^{13}\text{C}$  estimates associated with foraging at the APF ( $-21.2\text{‰}$ ) and STF ( $-18.3\text{‰}$ ), which are derived from tracked Wandering Albatrosses (Jaeger *et al.* 2010). The SAZ was defined as the waters bound to the north by the STF and to the south by the APF, and the Subtropical Zone (STZ) and Antarctic Zone (AZ) as the waters to the north of the STF and to the south of the APF, respectively. Separate linear mixed-effects models (LMMs) were constructed with feather  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values as response variables via the *lme4* package in R (Bates *et al.* 2015). Predictor variables were

sampling year, sex, age (12–37 years), previous breeding outcome and all two-way interactions. Grey-headed Albatrosses are predominantly biennial breeders; however, a minority attempt to breed annually (Ryan *et al.* 2007) and hence birds were grouped according to their breeding outcomes (successful, failed or deferred) in the 2 years prior to sampling. Individual identity was included as a random effect. All possible models were ranked using the Akaike information criterion adjusted for small sample sizes ( $AIC_C$ ) and models within 2  $AIC_C$  units of the top model ( $\leq 2 AIC_C$ ) were considered equally plausible (Burnham & Anderson 2002). Repeatabilities (intraclass correlation coefficient) in  $\delta^{13}C$  and  $\delta^{15}N$  values among different feathers of the same individual were calculated via the *rptR* package in R (Stoffel *et al.* 2017). Repeatability (range: 0–1) is calculated as the between-individual variance divided by the between- and within-individual variances (Carneiro *et al.* 2017) and our values are adjusted repeatabilities ( $R_{adj}$ ), as we controlled for predictor variables retained in the minimum adequate models (Nakagawa & Schielzeth 2010).

The isotopic niches of males and females were compared using the Stable Isotope Bayesian Ellipses in R package (*SIBER*; Jackson *et al.* 2011). Standard Ellipse Areas corrected for small sample sizes ( $SEA_C$ ), which represent the core isotopic niche, and 95% ellipse areas were calculated for each sex. Percentage overlaps in  $SEA_C$  and 95% ellipse areas were used to quantify sex differences in the isotopic niche. Bayesian standard ellipse areas ( $SEA_B$ ) are provided as unbiased estimates of core isotopic niche areas (presented as modes and 95% credibility intervals).

Finally, ordinal logistic regression was used to assess carry-over effects of stable isotope values on the subsequent breeding outcome using the *ordinal* package in R (Christensen 2019). The breeding outcome response variable had a natural ordering (failed at incubation ( $n = 23$ ); chick hatched but failed to fledge ( $n = 108$ ); chick fledged ( $n = 63$ )); all birds bred in the year that they were sampled. Two models were tested using the following predictors: (1) mean  $\delta^{15}N$  and  $\delta^{13}C$  values of individual birds and (2) variance of  $\delta^{15}N$  and  $\delta^{13}C$  values (normalized to the range: 0–1) based on the three feathers analysed for each individual. These models allowed us to test whether the absolute values or the variability is important. All intrinsic effects and sampling year were included as covariates.

Analyses were conducted using R version 3.4.4 (R Core Team 2019) and significance was set at  $\alpha = 0.05$ .

## RESULTS

### Intraspecific variation

A total of 582 isotopic measurements were made on body feathers of 194 adults; mean ( $\pm$ sd) stable isotope values ( $\delta^{13}C$ ,  $\delta^{15}N$ ) are presented in Table 1. Grey-headed Albatross feathers exhibited high variability in  $\delta^{13}C$  (range:  $-24.6$  to  $-17.1\%$ ) and  $\delta^{15}N$  values ( $6.7$ – $14.8\%$ ) (Fig. S1). According to the  $\delta^{13}C$  values, individuals foraged across a range of oceanic habitats during the non-breeding period, corresponding mostly to the SAZ and to a lesser extent to the AZ and STZ (Fig. 2; Table 2). The most parsimonious LMM ( $\Delta AIC_C = 0.0$ ) explaining  $\delta^{15}N$  values included sex as a fixed effect ( $F_{1,191} = 7.2$ ,  $P < 0.01$ ), reflecting higher  $\delta^{15}N$  values in males than in females (Fig. 2; Table 1). Models including other predictor variables (sampling year, age, breeding history) had less support ( $> 2 AIC_C$ ) (Table S1). The most parsimonious LMM explaining  $\delta^{13}C$  values also included sex as a fixed effect ( $F_{1,191} = 3.2$ ,  $P < 0.01$ ), reflecting lower  $\delta^{13}C$  in males than in females (Fig. 2; Tables 1 and 2); however, the null model and a model containing sampling year were equally competitive (Table S1). Models including other predictor variables (age, breeding history) received less support ( $> 2 AIC_C$ ) and the  $\delta^{13}C$  and  $\delta^{15}N$  values of younger individuals did not differ from those of older individuals (Table 3). Repeatability among feathers of the same individual was moderate in  $\delta^{13}C$  values ( $R_{adj} = 0.43 \pm 0.04$ , 95% CI 0.37–0.49,  $P < 0.001$ ) and low in  $\delta^{15}N$  values ( $R_{adj} = 0.15 \pm 0.05$ , 95% CI 0.07–0.24,  $P < 0.001$ ). Isotopic niches of males and females were similar in size (Table 1), with a 62% overlap in  $SEA_C$  and 80% overlap for the 95% ellipse areas (Fig. 2).

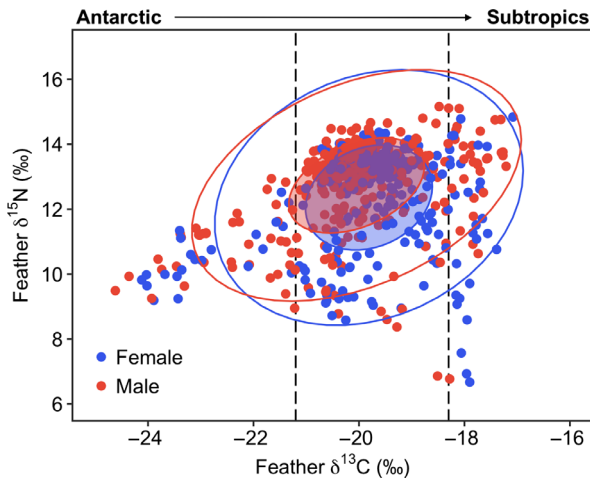
### Potential carry-over effects

Neither the mean (ordinal regression:  $\chi^2 = 0.10$ ,  $P = 0.75$ ) nor normalized variance in  $\delta^{13}C$  values ( $\chi^2 = 1.3$ ,  $P = 0.26$ ) had a significant effect on subsequent breeding outcome, and neither did the mean ( $\chi^2 = 0.53$ ,  $P = 0.61$ ) nor normalized variance in  $\delta^{15}N$  values ( $\chi^2 = 0.20$ ,  $P = 0.53$ ).  $\delta^{13}C$

**Table 1.** Mean ( $\pm$  sd) stable isotope values ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of three body feathers sampled from individual male and female Grey-headed Albatrosses *Thalassarche chrysostoma* at Bird Island, South Georgia (2013/14–2015/16).

Sex	<i>n</i>	$\delta^{13}\text{C}$ (‰)	$\delta^{15}\text{N}$ (‰)	SEA <sub>C</sub> (‰ <sup>2</sup> )	SEA <sub>B</sub> (‰ <sup>2</sup> )
Males	103	$-20.1 \pm 1.2$	$12.7 \pm 1.5$	5.32	5.32 (4.76, 5.94)
Females	91	$-19.8 \pm 1.2$	$12.3 \pm 1.6$	5.84	5.85 (5.19, 6.54)

SEA<sub>C</sub> is the standard ellipse area corrected for small sample size, and SEA<sub>B</sub> is the Bayesian estimate of the standard ellipse (modes with 95% credibility intervals in parentheses).

**Figure 2.** Stable isotope values ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of individual body feathers from adult Grey-headed Albatrosses *Thalassarche chrysostoma* at Bird Island, South Georgia (2013/14–2015/16). Vertical lines are  $\delta^{13}\text{C}$  estimates of the Antarctic Polar Front ( $-21.2\text{‰}$ ) and Subtropical Front ( $-18.3\text{‰}$ ), separating the Antarctic Zone, Subantarctic Zone and Subtropical Zone (Jaeger *et al.* 2010). Standard ellipse areas corrected for small sample sizes (SEA<sub>C</sub>; shaded) and 95% ellipse areas are shown.

values and  $\delta^{15}\text{N}$  (means and variances) among birds that failed at the incubation or chick stage were therefore similar to those of individuals that successfully fledged their chick.

## DISCUSSION

Stable isotope values of adult feathers are considered to be effective proxies of habitat use and trophic level of albatrosses during the comparatively understudied non-breeding season (Cherel *et al.* 2000, 2013, Phillips *et al.* 2009). Albatrosses forage over marine isoscapes (reflecting broad-scale isotopic variation), and spatial variation in  $\delta^{13}\text{C}$  is reflected in their tissues (Quillfeldt *et al.* 2005, Cherel & Hobson 2007). The specific threshold

**Table 2.** Numbers of Grey-headed Albatross *Thalassarche chrysostoma* body feathers with  $\delta^{13}\text{C}$  values corresponding to  $\delta^{13}\text{C}$  estimates of foraging in the Antarctic Zone, Subantarctic Zone and Subtropical Zone (Jaeger *et al.* 2010).

	Antarctic Zone	Subantarctic Zone	Subtropical Zone
Females	22 (8.0)	228 (82.6)	26 (9.4)
Males	38 (12.4)	238 (77.8)	30 (9.8)
Total	60 (10.3)	466 (80.1)	56 (9.6)

Percentages are in parentheses. All sampled birds were from Bird Island, South Georgia (2013/14–2015/16).

**Table 3.** Mean ( $\pm$  sd) stable isotope values ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of three body feathers sampled from younger, mid-aged and older Grey-headed Albatrosses *Thalassarche chrysostoma* at Bird Island, South Georgia (2013/14–2015/16). Age classes are delineated following Catry *et al.* (2006).

Age class	<i>n</i>	$\delta^{13}\text{C}$ (‰) <sup>a</sup>	$\delta^{15}\text{N}$ (‰) <sup>b</sup>
Younger ( $\leq 14$ years)	13	$-20.2 \pm 0.7$	$12.6 \pm 0.9$
Mid-aged (15–27 years)	119	$-20.0 \pm 0.9$	$12.5 \pm 1.0$
Older ( $\geq 28$ years)	62	$-19.8 \pm 1.0$	$12.7 \pm 1.1$

<sup>a</sup>No significant differences among age classes (ANOVA,  $F_{2,192} = 1.43$ ,  $P = 0.24$ ). <sup>b</sup>No significant differences among age classes (ANOVA,  $F_{2,192} = 0.09$ ,  $P = 0.92$ ).

$\delta^{13}\text{C}$  values that we used to assign moulting location to north or south of the APF and STF were derived from tracked Wandering Albatrosses in the Indian Ocean (Jaeger *et al.* 2010). However, the paths of the main oceanographic fronts in the Southern Ocean can be highly variable between years (Moore *et al.* 1997) and stable isotope values are therefore broadly indicative of water masses rather than latitude per se. Allowing for some uncertainty, feather  $\delta^{13}\text{C}$  values indicate that Grey-headed Albatrosses sampled at the Bird Island colony in 2013/14–2015/16 foraged

predominantly in the SAZ, and to a lesser extent in the STZ and AZ, during the previous non-breeding period. This is consistent with stable isotope data from other populations of this species (Cherel *et al.* 2013) and with geolocator data from a much smaller sample of individuals tracked previously from South Georgia (Croxall *et al.* 2005, Clay *et al.* 2016). Hence, our first a priori hypothesis that most individuals spent the non-breeding period in the SAZ is supported.

### Drivers of variation in habitat use and trophic level

Our study highlights considerable intraspecific variation in the habitat use ( $\delta^{13}\text{C}$ ) and trophic level ( $\delta^{15}\text{N}$ ) of Grey-headed Albatrosses during the non-breeding period. Sexual segregation and other sex differences in foraging ecology are well-documented among seabirds, including albatrosses (Phillips *et al.* 2004, 2011, Weimerskirch *et al.* 2014, Froy *et al.* 2015). In our analyses, sex was the best predictor of feather  $\delta^{13}\text{C}$  and  $\delta^{15}\text{N}$  values, although differences were small – mean feather  $\delta^{13}\text{C}$  values were slightly lower and  $\delta^{15}\text{N}$  values slightly higher in males than in females. Moreover, our analyses of the isotopic niche show high, but not complete, overlap between sexes. These differences indicate that males forage at higher latitudes and to a greater extent on higher trophic level prey, hence providing some support for our second a priori prediction. The aforementioned geolocator data showed that males foraged at slightly higher latitudes (by *c.*  $1^\circ$ ), and core areas but not overall distributions were segregated to some extent from females during the non-breeding summer only (Clay *et al.* 2016). Moreover, a previous stable isotope study, albeit with a much reduced sample size, also found that  $\delta^{13}\text{C}$  values in body feathers of males were lower than those of females (Phillips *et al.* 2009). Male Grey-headed Albatrosses are 15% heavier, with overall wing area and wing loading greater by 5 and 10%, respectively, than females (Phillips *et al.* 2004). This may confer a functional role in flight performance (Shaffer *et al.* 2001), with males perhaps better able to take advantage of the stronger winds at higher latitudes with associated flight cost reductions (Phillips *et al.* 2004, Jaeger *et al.* 2014). The slightly higher  $\delta^{15}\text{N}$  values could indicate that males consume a greater proportion of higher trophic level prey during the non-breeding period, or larger

individuals rather than different species, as size-related increases in  $\delta^{15}\text{N}$  values in fish, squid and crustaceans are often apparent within taxa (Schmidt *et al.* 2003). Male Grey-headed Albatrosses have a longer and deeper bill than females (Phillips *et al.* 2004), which could conceivably enable them to manipulate larger prey. Given the difficulties in obtaining samples, no complete conventional diet studies (i.e. for all prey taxa) exist outside the breeding period. Despite differing foraging distributions in incubation (Phillips *et al.* 2004), there is no evidence for consistent differences between sexes in activity patterns in Grey-headed Albatrosses during the breeding season (Phalan *et al.* 2007). Nor do activity patterns differ between sexes in the closely related Black-browed Albatross *Thalassarche melanophris* or in Wandering Albatrosses during the non-breeding season (Mackley *et al.* 2010). Spatial variation in  $\delta^{15}\text{N}$  baselines can confound interpretation; however, that is very unlikely to be problematic in our comparison, as  $\delta^{15}\text{N}$  values actually decrease with latitude in the Southern Ocean (Jaeger *et al.* 2010) and males both had higher  $\delta^{15}\text{N}$  values and foraged at higher latitudes according to  $\delta^{13}\text{C}$ .

Our analyses provided no support for our a priori prediction relating to age-related variation in foraging (Table 3). This contrasts with previous studies of Wandering Albatrosses. Tracking at the Crozet Islands showed that older males foraged further south with increasing age during the breeding season (Lecomte *et al.* 2010), although this was not found at South Georgia (Froy *et al.* 2015). An age-related decrease in  $\delta^{13}\text{C}$  values in body feathers—representing the non-breeding period—was observed in Wandering Albatrosses at South Georgia (Clay *et al.* 2018) and at the Crozet Islands (Jaeger *et al.* 2014). Grey-headed Albatrosses show distinct age-specific habitat preferences in terms of sea surface temperature during the breeding season (Frankish *et al.* 2020) and older birds ( $\geq 35$  years) took longer trips and had lower daily mass gains compared with mid-aged birds ( $\leq 28$  years) (Catry *et al.* 2006). However, our results suggest that if there are age-related changes in at-sea activity patterns, foraging habitat or trophic level in Grey-headed Albatrosses at South Georgia during the non-breeding period, then they are at fine scales and not evident from stable isotope values. Additionally, there is the caveat that most studies (including the present study) are cross-sectional and hence the possibility

of selective mortality of particular phenotypes cannot be excluded except by carrying out longitudinal studies.

### Variation among feathers

Jaeger *et al.* (2009) noted that measuring stable isotopes in multiple feathers from the same individual could provide insights into within-individual variation, which contrasts with the conventional procedure of pooling multiple feathers per individual. After accounting for sex effects, repeatability among feathers was moderate in  $\delta^{13}\text{C}$  and low in  $\delta^{15}\text{N}$ , but significant in both cases, for which there are two possible explanations. First, repeatability may be a consequence of sampling feathers that were regrown over broadly the same period. However, this is unlikely given that body feathers are most likely replaced gradually over the non-breeding period (see Methods). A second alternative explanation is that individuals showed some consistency in their habitat use and, to a lesser extent, trophic level during the non-breeding period. Given that seabirds are dependent on resources that are patchily distributed but predictable at large spatial scales (Weimerskirch 2007), a degree of consistency in foraging areas would be unsurprising. Moreover, from a limited number of tracked individuals, Croxall *et al.* (2005) found consistency in the habitats used by Grey-headed Albatrosses in successive winters during the non-breeding period. Finally, chick-rearing Grey-headed Albatrosses show individual foraging site fidelity and specialization in habitat use in terms of sea surface temperature, eddy kinetic energy and water depth (Bonnet-Lebrun *et al.* 2018).

### Carry-over effects

Behaviour or conditions experienced during the non-breeding period have an influence on subsequent breeding outcome in some albatrosses (Crossin *et al.* 2013, Clay *et al.* 2018). Grey-headed Albatrosses lay a single egg clutch with no replacement, and consistently successful birds at South Georgia arrive earlier at the colony, have shorter incubation shifts, and hatch larger chicks with higher growth rates compared with less successful birds (Cobley *et al.* 1998). Diet in the preceding non-breeding period may influence body condition, which can have consequences for subsequent reproduction (Sorensen *et al.* 2009).

Nonetheless, in our analyses, no significant relationships were found between mean feather stable isotope values, or variance in such values, and breeding outcome. Moreover, previous breeding outcome did not explain stable isotope values. Any potential relationships between stable isotopes, reflecting distribution and diet, would probably be mediated through variation in body condition on arrival at the colony. The decision to breed in Grey-headed Albatrosses at South Georgia is influenced by body condition (Crossin *et al.* 2013). All birds in our study were sampled as breeders, from which we can infer that they returned to the colony in relatively good condition.

### CONCLUSION

Rather less is known about the foraging ecology of albatrosses during the non-breeding compared with the breeding season. A novel insight from our study is the high level of variability in habitat use and trophic levels of non-breeding Grey-headed Albatrosses from South Georgia. Grey-headed Albatrosses are therefore population-level generalists during the non-breeding periods and utilize a range of oceanic habitats, although they mainly target the SAZ according to feather  $\delta^{13}\text{C}$  values; this is confirmed by geolocator data from previous tracking studies of this population. Sex differences were subtle, but significant, and the overlap in the core isotopic niche was high but not complete. Neither age nor previous breeding outcome influenced stable isotope values. Future research on albatross foraging ecology, particularly age-related changes, would benefit from longitudinal studies, and stable isotope studies would benefit from increased understanding of moulting patterns. We also found no evidence of carry-over effects of non-breeding diet or distribution on subsequent breeding outcome and it would be useful to examine carry-over effects on birds observed as non-breeders at the colony, which are likely to vary more in terms of physiological condition.

The authors thank all field assistants involved in collecting albatross feather samples at Bird Island. Constructive comments from two anonymous reviewers substantially improved this work. W.F.M. is supported by an NERC GW4+ Doctoral Training Partnership studentship from the Natural Environment Research Council (NE/L002434/1). Stable isotope analyses were funded by the NERC Life Sciences Mass Spectrometry Facility (grant number EK311-12/18). This work represents a

contribution to the Ecosystems component of the British Antarctic Survey Polar Science for Planet Earth Programme, funded by the Natural Environment Research Council.

## AUTHOR CONTRIBUTION

**William F. Mills:** Conceptualization (equal); Formal analysis (lead); Investigation (equal); Methodology (equal); Visualization (lead); Writing-original draft (lead). **Rona A. R. McGill:** Investigation (equal); Methodology (equal); Writing-review & editing (equal). **Yves Cherel:** Supervision (equal); Writing-review & editing (equal). **Stephen C. Votier:** Supervision (equal); Writing-review & editing (equal). **Richard A. Phillips:** Conceptualization (equal); Resources (lead); Supervision (equal); Writing-review & editing (equal).

## Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

## REFERENCES

- Bates, D., Maechler, M., Bolker, B. & Walker, S. 2015. Fitting linear mixed-effects models using lme4. *J. Stat. Softw.* **67**: 1–48.
- Battam, H., Richardson, M., Watson, A.W.T. & Buttemer, W.A. 2010. Chemical composition and tissue energy density of the cuttlefish (*Sepia apama*) and its assimilation efficiency by *Diomedea* albatrosses. *J. Comp. Physiol. B.* **180**: 1247–1255.
- Bearhop, S., Waldron, S., Votier, S.C. & Furness, R.W. 2002. Factors that influence assimilation rates and fractionation of nitrogen and carbon stable isotopes in avian blood and feathers. *Physiol. Biochem. Zool.* **75**: 451–458.
- Bearhop, S., Phillips, R.A., McGill, R.A.R., Cherel, Y., Dawson, D.A. & Croxall, J.P. 2006. Stable isotopes indicate sex-specific and long-term individual foraging specialisation in diving seabirds. *Mar. Ecol. Prog. Ser.* **311**: 157–164.
- Bolnick, D.I., Svanbäck, R., Fordyce, J.A., Yang, L.H., Davis, J.M., Hulsey, C.D. & Forister, M.L. 2003. The ecology of individuals: incidence and implications of individual specialization. *Am. Nat.* **161**: 1–28.
- Bonnet-Lebrun, A.S., Phillips, R.A., Manica, A. & Rodrigues, A.S.L. 2018. Quantifying individual specialization using tracking data: a case study on two species of albatrosses. *Mar. Biol.* **165**: 152.
- Burnham, K.P. & Anderson, D.R. 2002. *Model Selection and Inference: A Practical Information-theoretic Approach*. New York, NY: Springer.
- Carneiro, A.P.B., Bonnet-Lebrun, A.S., Manica, A., Staniland, I.J. & Phillips, R.A. 2017. Methods for detecting and quantifying individual specialisation in movement and foraging strategies of marine predators. *Mar. Ecol. Prog. Ser.* **578**: 151–166.
- Catry, P., Phillips, R.A., Phalan, B. & Croxall, J.P. 2006. Senescence effects in an extremely long-lived bird: the Grey-headed Albatross *Thalassarche chrysostoma*. *Proc. R. Soc. B. Biol. Sci.* **273**: 1625–1630.
- Catry, P., Poisbleau, M., Lecoq, M. & Phillips, R.A. 2013. Differences in the timing and extent of annual moult of Black-browed Albatrosses *Thalassarche melanophris* living in contrasting environments. *Polar Biol.* **36**: 837–842.
- Cherel, Y. & Hobson, K.A. 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.A. & Weimerskirch, H. 2000. Using stable-isotope analysis of feathers to distinguish moulting and breeding origins of seabirds. *Oecologia* **122**: 155–162.
- Cherel, Y., Jaeger, A., Alderman, R., Jaquemet, S., Richard, P., Wanless, R.M., Phillips, R.A. & Thompson, D.R. 2013. A comprehensive isotopic investigation of habitat preferences in nonbreeding albatrosses from the Southern Ocean. *Ecography* **36**: 277–286.
- Christensen, R.H.B. 2019. *Ordinal – Regression Models for Ordinal Data*. R package version 2019.4-25. <http://www.cran.r-project.org/package=ordinal/>. Accessed January 1, 2020.
- Clay, T.A., Manica, A., Ryan, P.G., Silk, J.R.D., Croxall, J.P., Ireland, L. & Phillips, R.A. 2016. Proximate drivers of spatial segregation in non-breeding albatrosses. *Sci. Rep.* **6**: 29932.
- Clay, T.A., Pearmain, E.J., McGill, R.A.R., Manica, A. & Phillips, R.A. 2018. Age-related variation in non-breeding foraging behaviour and carry-over effects on fitness in an extremely long-lived bird. *Funct. Ecol.* **32**: 1832–1846.
- Cobley, N.D., Croxall, J.P. & Prince, P.A. 1998. Individual quality and reproductive performance in the Grey-headed Albatross *Diomedea chrysostoma*. *Ibis* **140**: 315–322.
- Crossin, G.T., Phillips, R.A., Wynne-Edwards, K.E. & Williams, T.D. 2013. Postmigratory body condition and ovarian steroid production predict breeding decisions by female Gray-headed Albatrosses. *Physiol. Biochem. Zool.* **86**: 761–768.
- Croxall, J.P., Silk, J.R.D., Phillips, R.A., Afanasyev, V. & Briggs, D.R. 2005. Global circumnavigations: tracking year-round ranges of nonbreeding albatrosses. *Science* **307**: 249–250.
- De Felipe, F., Reyes-González, J.M., Militão, T., Neves, V.C., Bried, J., Oro, D., Ramos, R. & González-Solís, J. 2019. Does sexual segregation occur during the nonbreeding period? A comparative analysis in spatial and feeding ecology of three *Calonectris* shearwaters. *Ecol. Evol.* **9**: 10145–10162.
- DeNiro, M. & Epstein, S. 1981. Influence of diet on the distribution of nitrogen isotopes in animals. *Geochim. Cosmochim. Acta* **45**: 341–351.
- Frankish, C.K., Manica, A. & Phillips, R.A. 2020. Effects of age on foraging behavior in two closely related albatross species. *Mov. Ecol.* **8**: 7.
- Fridolfsson, A. & Ellegren, H. 1999. A simple and universal method for molecular sexing of non-ratite birds. *J. Avian Biol.* **30**: 116–121.
- Froy, H., Lewis, S., Catry, P., Bishop, C.M., Forster, I.P., Fukuda, A., Higuchi, H., Phalan, B., Xavier, J.C., Nussey, D.H. & Phillips, R.A. 2015. Age-related variation in foraging



- behaviour in the wandering albatross at South Georgia: no evidence for senescence. *PLoS One* **10**: e0116415.
- Froy, H., Lewis, S., Nussey, D.H., Wood, A.G. & Phillips, R.A. 2017. Contrasting drivers of reproductive ageing in albatrosses. *J. Anim. Ecol.* **86**: 1022–1032.
- Hobson, K.A. & Clark, R.G. 1992. Assessing avian diets using stable isotopes I: turnover of  $^{13}\text{C}$  in tissues. *Condor* **94**: 181–188.
- Jackson, A.L., Inger, R., Parnell, A.C. & 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., Blanchard, P., Richard, P. & Cherel, Y. 2009. Using carbon and nitrogen isotopic values of body feathers to infer inter- and intra-individual variations of seabird feeding ecology during moult. *Mar. Biol.* **156**: 1233–1240.
- Jaeger, A., Lecomte, V.J., Weimerskirch, H., Richard, P. & Cherel, Y. 2010. Seabird satellite tracking validates the use of latitudinal isoscapes to depict predators' foraging areas in the Southern Ocean. *Rapid Commun. Mass Spectrom.* **24**: 3456–3460.
- Jaeger, A., Goutte, A., Lecomte, V.J., Richard, P., Chastel, O., Barbraud, C., Weimerskirch, H. & Cherel, Y. 2014. Age, sex, and breeding status shape a complex foraging pattern in an extremely long-lived seabird. *Ecology* **95**: 2324–2333.
- Lamacchia, P., Madrid, E.A. & Mariano-Jelicich, R. 2019. Intraspecific variability in isotopic composition of a monomorphic seabird, the Common Tern (*Sterna hirundo*), at wintering grounds. *Emu* **119**: 176–185.
- Lecomte, V.J., Sorci, G., Cornet, S., Jaeger, A., Faivre, B., Arnoux, E., Gaillard, M., Trouve, C., Besson, D., Chastel, O. & Weimerskirch, H. 2010. Patterns of aging in the long-lived wandering albatross. *Proc. Natl Acad. Sci. USA*. **107**: 6370–6375.
- Mackley, E.K., Phillips, R.A., Silk, J.R.D., Wakefield, E.D., Afanasyev, V., Fox, J.W. & Furness, R.W. 2010. Free as a bird? Activity patterns of albatrosses during the nonbreeding period. *Mar. Ecol. Prog. Ser.* **406**: 291–303.
- Mancini, P.L., Bond, A.L., Hobson, K.A., Duarte, L.S. & Bugoni, L. 2013. Foraging segregation in tropical and polar seabirds: testing the Intersexual Competition Hypothesis. *J. Exp. Mar. Biol. Ecol.* **449**: 186–193.
- Mills, W.F., Xavier, J.C., Bearhop, S., Cherel, Y., Votier, S.C., Waluda, C.M. & Phillips, R.A. 2020. Long-term trends in albatross diets in relation to prey availability and breeding success. *Mar. Biol.* **167**: 29.
- Moore, J., Abbott, M. & Richman, J. 1997. Variability in the location of the Antarctic Polar Front (90°–20°W) from satellite sea surface temperature data. *J. Geophys. Res.* **102**: 27825–27833.
- Nakagawa, S. & Schielzeth, H. 2010. Repeatability for Gaussian and non-Gaussian data: a practical guide for biologists. *Biol. Rev.* **85**: 935–956.
- Pardo, D., Forcada, J., Wood, A.G., Tuck, G.N., Ireland, L., Pradel, R., Croxall, J.P. & Phillips, R.A. 2017. Additive effects of climate and fisheries drive ongoing declines in multiple albatross species. *Proc. Natl Acad. Sci. USA*. **114**: E10829–E10837.
- Phalan, B., Phillips, R.A., Silk, J.R.D., Afanasyev, V., Fukuda, A., Fox, J., Catry, P., Higuchi, H. & Croxall, J.P. 2007. Foraging behaviour of four albatross species by night and day. *Mar. Ecol. Prog. Ser.* **340**: 271–286.
- Phillips, R.A., Silk, J.R.D., Phalan, B., Catry, P. & Croxall, J.P. 2004. Seasonal sexual segregation in two *Thalassarche* albatross species competitive exclusion reproductive role specialization or foraging niche divergence. *Proc. R. Soc. B. Biol. Sci.* **271**: 1283–1291.
- Phillips, R.A., Bearhop, S., McGill, R.A.R. & Dawson, D.A. 2009. Stable isotopes reveal individual variation in migration strategies and habitat preferences in a suite of seabirds during the nonbreeding period. *Oecologia* **160**: 795–806.
- Phillips, R.A., McGill, R.A.R., Dawson, D.A. & Bearhop, S. 2011. Sexual segregation in distribution, diet and trophic level of seabirds: insights from stable isotope analysis. *Mar. Biol.* **158**: 2199–2208.
- Phillips, R.A., Lewis, S., González-Solís, J. & Daunt, F. 2017. Causes and consequences of individual variability and specialization in foraging and migration strategies of seabirds. *Mar. Ecol. Prog. Ser.* **578**: 117–150.
- Poncet, S., Wolfaardt, A.C., Black, A., Browning, S., Lawton, K., Lee, J., Passfield, K., Strange, G. & Phillips, R.A. 2017. Recent trends in numbers of Wandering (*Diomedea exulans*), Black-browed (*Thalassarche melanophris*) and Grey-headed (*T. chrysostoma*) albatrosses breeding at South Georgia. *Polar Biol.* **40**: 1347–1358.
- Prince, P.A., Rodwell, S., Jones, M. & Rothery, P. 1993. Molt in Black-browed and Grey-headed Albatrosses *Diomedea melanophris* and *D. chrysostoma*. *Ibis* **135**: 121–131.
- Prince, P.A., Rothery, P., Croxall, J.P. & Wood, A.G. 1994. Population dynamics of Black-browed and Grey-headed Albatrosses *Diomedea melanophris* and *D. chrysostoma* at Bird Island, South Georgia. *Ibis* **136**: 50–71.
- Quillfeldt, P., McGill, R.A.R. & Furness, R.W. 2005. Diet and foraging areas of Southern Ocean seabirds and their prey inferred from stable isotopes: review and case study of Wilson's Storm-petrel. *Mar. Ecol. Prog. Ser.* **295**: 295–304.
- Quillfeldt, P., Masello, J.F., McGill, R.A.R., Adams, M. & Furness, R.W. 2010. Moving polewards in winter: a recent change in the migratory strategy of a pelagic seabird? *Front. Zool.* **7**: 15.
- R Core Team. 2019. *R: A Language and Environment for Statistical Computing*. Vienna: R Foundation for Statistical Computing. <https://www.R-project.org/>.
- Ryan, P.G., Phillips, R.A., Nel, D.C. & Wood, A.G. 2007. Breeding frequency in Grey-headed Albatrosses *Thalassarche chrysostoma*. *Ibis* **149**: 45–52.
- Schmidt, K., Atkinson, A., Stübing, D., McClelland, J.W., Montoya, J.P. & Voss, M. 2003. Trophic relationships among Southern Ocean copepods and krill: some uses and limitations of a stable isotope approach. *Limnol. Oceanogr.* **48**: 277–289.
- Shaffer, S.A., Weimerskirch, H. & Costa, D.P. 2001. Functional significance of sexual dimorphism in Wandering Albatrosses, *Diomedea exulans*. *Funct. Ecol.* **15**: 203–210.
- Sorensen, M.C., Hipfner, J.M., Kyser, T.K. & Norris, D.R. 2009. Carry-over effects in a Pacific seabird: stable isotope evidence that pre-breeding diet quality influences reproductive success. *J. Anim. Ecol.* **78**: 460–467.
- Stoffel, M.A., Nakagawa, S. & Schielzeth, H. 2017. rptR: Repeatability estimation and variance decomposition by generalized linear mixed-effects models. *Methods Ecol. Evol.* **8**: 1639–1644.
- Votier, S.C., Bearhop, S., Witt, M.J., Inger, R., Thompson, D. & Newton, J. 2010. Individual responses of seabirds to commercial fisheries revealed using GPS tracking, stable

isotopes and vessel monitoring systems. *J. Appl. Ecol.* **47**: 487–497.

- Votier, S.C., Fayet, A.L., Bearhop, S., Bodey, T.W., Clark, B.L., Grecian, J., Guilford, T., Hamer, K.C., Jeglinski, J.W.E., Morgan, G., Wakefield, E. & Patrick, S.C.** 2017. Effects of age and reproductive status on individual foraging site fidelity in a long-lived marine predator. *Proc. R. Soc. B. Biol. Sci.* **284**: 20171068.
- Weimerskirch, H.** 2007. Are seabirds foraging for unpredictable resources?. *Deep-Sea Res. II* **54**: 211–223.
- Weimerskirch, H., Cherel, Y., Delord, K., Jaeger, A., Patrick, S.C. & Riotte-Lambert, L.** 2014. Lifetime foraging patterns of the Wandering Albatross: life on the move!. *J. Exp. Mar. Biol. Ecol.* **450**: 68–78.

Received 10 January 2020;  
revision accepted 4 August 2020.  
Associate Editor: Morten Frederiksen.

## SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.

**Table S1.** Model selection for factors explaining variation in stable isotope values ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of Grey-headed Albatross *Thalassarche chrysostoma* body feathers at Bird Island, South Georgia (2013/14–2015/16).

**Figure S1.** Mean ( $\pm$  standard deviation) stable isotope values ( $\delta^{13}\text{C}$ ,  $\delta^{15}\text{N}$ ) of body feathers from adult Grey-headed Albatrosses *Thalassarche chrysostoma* at Bird Island, South Georgia (2013/14–2015/16).