

Supplementary Material

Contents

1 Time incorporated in otolith samples	2
1.1 <i>Electrona antarctica</i>	2
1.2 <i>Gymnoscopelus braueri</i>	3
1.3 <i>Krefftichthys anderssoni</i>	4
1.4 <i>Electrona carlsbergi</i>	5
1.5 <i>Protomyctophum bolini</i>	6
1.6 <i>Gymnoscopelus nicholsi</i>	7
2 Effect of otolith preparation method on C_{resp} and temperature	8
3 Effect life stage on C_{resp} values	11
4 Further investigations within species	14
4.1 Effect of year of capture within Species	14
4.2 Further investigations within <i>Protomyctophum bolini</i>	16
4.3 Further investigations within <i>Gymnoscopelus nicholsi</i>	20
5 Age estimations of study individuals from length	21
References.....	22

1 Time incorporated in otolith samples

To estimate the amount of time incorporated into each otolith sample, we prepared sagittal thin sections of representative otoliths for each species. We mounted otoliths in cyanoacrylate glue and polished each face using 30 μm aluminium oxide lapping paper. We photographed sections using a Wild Heerbrugg 3M microscope using transmitted light. Annuli were measured as a pair of hyaline and opaque bands, with the edge of an annulus taken as the edge of the opaque band. All images were analysed and annotated in ImageJ 1.52a.

1.1 *Electrona antarctica*

From a female, 71 mm SL (BAS_33). We estimated the age of the individual was 3.5 years. We estimated the amount of time incorporated in the otolith sample as 2 years.

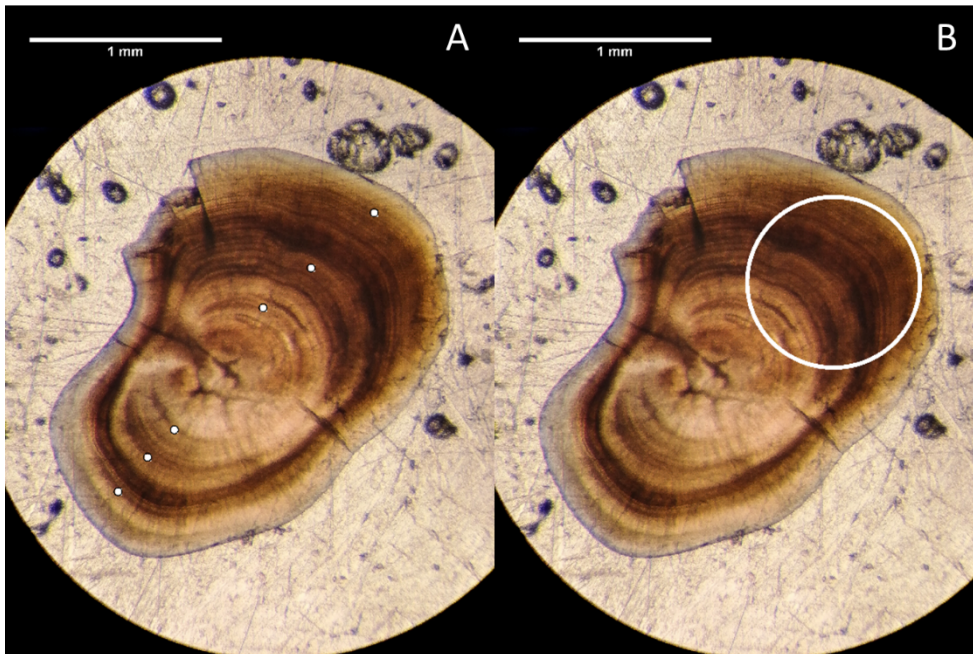


Figure S1: Section of an otolith from *Electrona antarctica* (BAS_33, female, 71 mm SL) taken at 40x magnification. White dots indicate estimated annuli (A), and the white circle indicates a representative sampling point with a cut width of 895 μm (B).

1.2 *Gymnoscopelus braueri*

From a male, 125 mm SL (BAS_94). We estimated the age of the individual was 6 years. We estimated the amount of time incorporated in the otolith sample as 3.5 years.

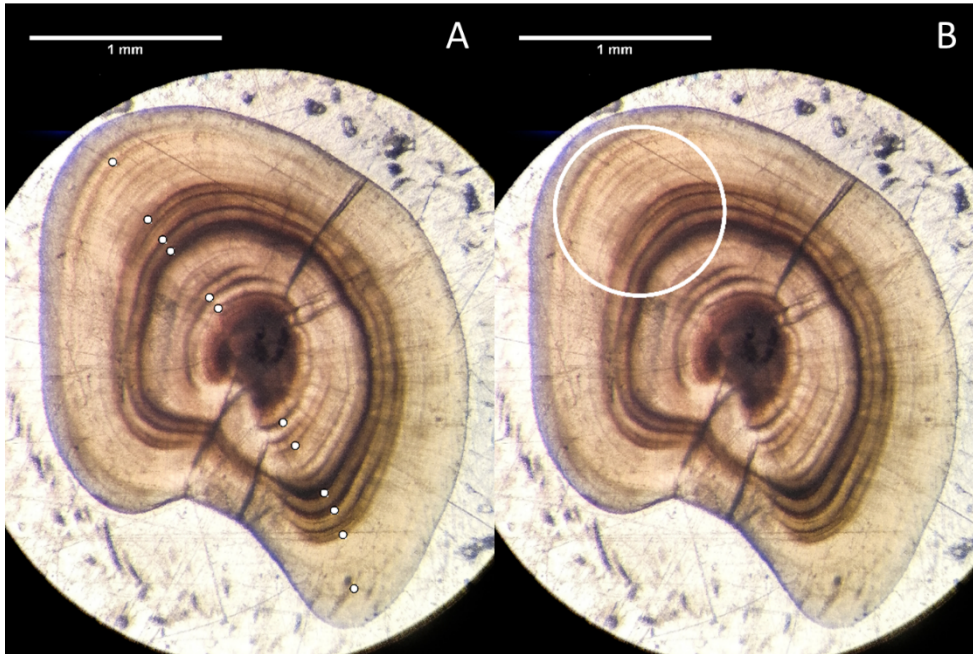


Figure S2: Section of an otolith from *Gymnoscopelus braueri* (BAS_94, male, 124 mm SL) taken at 40x magnification. White dots indicate estimated annuli (A), and the white circle indicates a representative sampling point with a cut width of 895 μm (B).

1.3 *Krefftichthys anderssoni*

From a female, 43 mm SL (not included in dataset). We estimated the age of the individual was 1.5 years. As *K. anderssoni* otoliths were all crushed, we estimated the amount of time incorporated in the otolith sample is also 1.5 years.

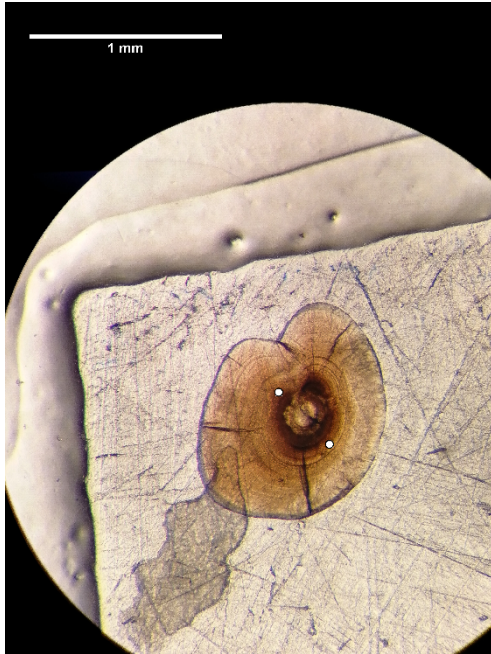


Figure S3: Section of an otolith from *Krefftichthys anderssoni* (female, 43 mm SL) taken at 40x magnification. White dots indicate estimated annuli.

1.4 *Electrona carlsbergi*

From an individual of undetermined sex and size (BAS_84). We estimated the age of the individual was 3 years. We estimated the amount of time incorporated in the otolith sample as 1 year.

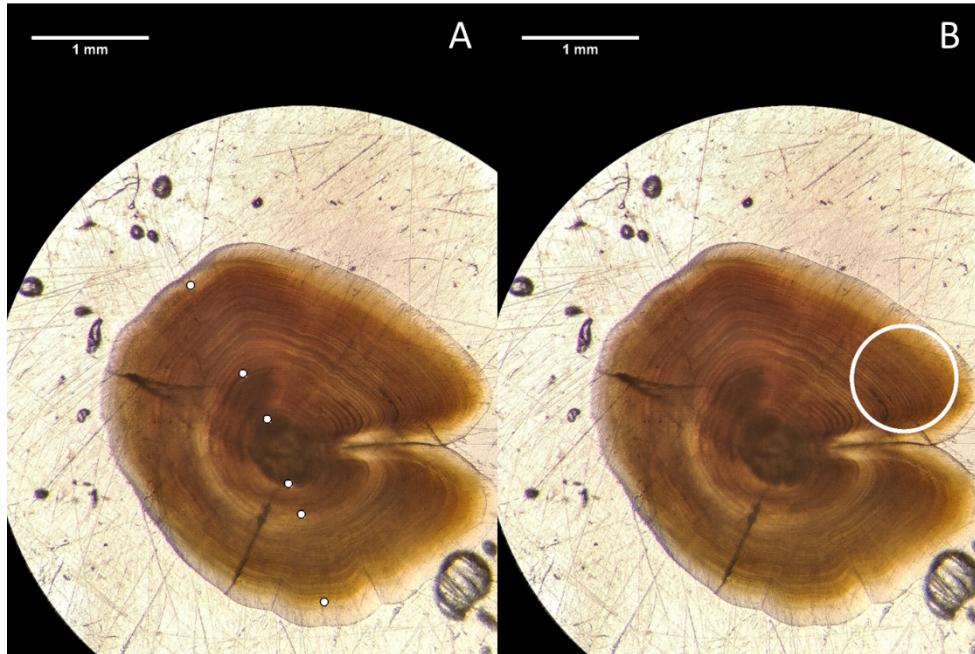


Figure S4: Section of an otolith from *Electrona carlsbergi* (BAS_84, undetermined sex and length) taken at 25x magnification. White dots indicate estimated annuli (A), and the white circle indicates a representative sampling point with a cut width of 895 μm (B).

1.5 *Protomyctophum bolini*

From a male, 44 mm SL (BAS_212). We estimated the age of the individual was 2.5 years. We estimated the amount of time incorporated in the otolith sample as 2 years.

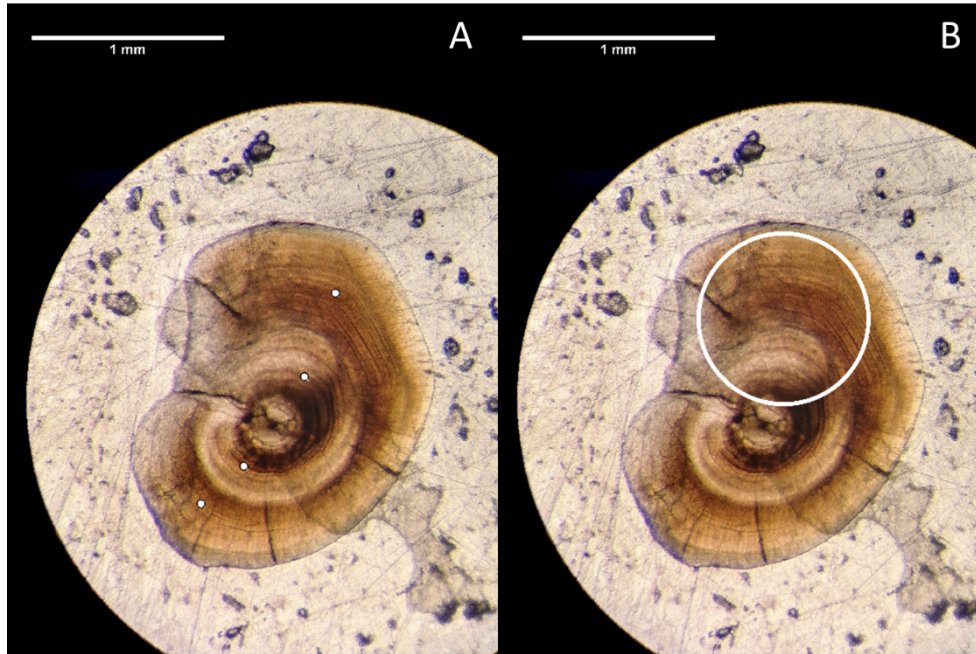


Figure S5: Section of an otolith from *Protomyctophum bolini* (BAS_212, male, 44 mm SL) taken at 40x magnification. White dots indicate estimated annuli (A), and the white circle indicates a representative sampling point with a cut width of 895 μm (B).

1.6 *Gymnoscopelus nicholsi*

From a male, 151 mm SL (BAS_122). We estimated the age of the individual was 5 years. We estimated the amount of time incorporated in the otolith sample as 2.5 years.

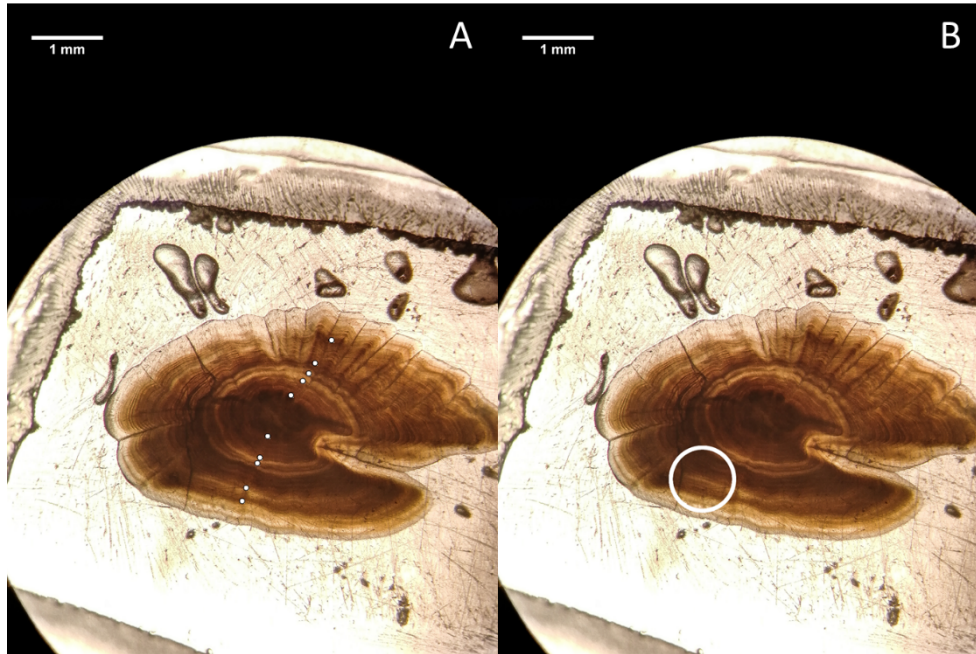


Figure S6: Section of an otolith from *Gymnoscopelus nicholsi* (BAS_122, male, 151 mm SL) taken at 16x magnification. White dots indicate estimated annuli (A), and the white circle indicates a representative sampling point with a cut width of 895 μm (B).

2 Effect of otolith preparation method on C_{resp} and temperature

To obtain powder for stable isotope analysis, we used two methods of preparation. Larger otoliths (>1mm diameter) were milled, sampling only the outer surface, while smaller otoliths (≤ 1 mm diameter) were crushed, sampling the whole otolith including the core. To test whether this had a significant impact on resulting C_{resp} values, we modelled C_{resp} values as a function of otolith preparation method, with species as a random factor:

$$C_{\text{resp}} = a + b_{\text{Prep}} \times \text{Prep} + a_{\text{VarSpecies}} \quad (\text{S1})$$

Where *Prep* is the method of preparation assigned to a dummy variable (milled = 0, crushed = 1) and b_{Prep} is the effect of crushing the otolith on C_{resp} , and $a_{\text{VarSpecies}}$ is the variable intercept for species. As with other models, we ran this in R version 4.0.5 (R Core Team 2021) with RStan version 2.21.2 (Stan Development Team 2020) using the rethinking package version 2.01 (McElreath 2020). We ran a single chain of 10000 iterations, 5000 warmup and a thinning parameter of one. We did the same for temperature, modifying equation S1 to swap C_{resp} for temperature (T):

$$T = a + b_{\text{Prep}} \times \text{Prep} + a_{\text{VarSpecies}} \quad (\text{S2})$$

Crushing did significantly increase C_{resp} values, indicated by b_{Prep} 95% HDPIs not overlapping zero (Figure S7A), however this significance was marginal; the value for b_{Prep} was 0.026 ± 0.019 . Additionally this model's diagnostics did show some concerning behaviour (Gelman-Rubin diagnostic >1.01 and low effective sample size), so should be treated with caution. There was no significant effect of preparation method on temperature (Figure S7B).

Most otoliths from *P. bolini* were milled, but two small otoliths were crushed. Therefore we ran the same model, without the variable intercept for species, to test for a difference in C_{resp} values or temperature between crushed and milled otoliths within *Protomyctophum bolini*:

$$C_{\text{resp}} = a + b_{\text{Prep}} \times \text{Prep} \quad (\text{S3})$$

$$T = a + b_{\text{Prep}} \times \text{Prep} \quad (\text{S4})$$

There was no significant effect of preparation method on C_{resp} values or temperature within this species (Figure S8).

Given the unbalanced design of these two models, it is unclear whether preparation method had a significant effect on C_{resp} values. Given that *Krefflichthys anderssoni* C_{resp} values were entirely estimated using crushed otolith samples, it is likely that *K. anderssoni* C_{resp} values are slightly higher due to this preparation method, and this should be born in mind when interpreting our results. As only two *P. bolini* otoliths were crushed, this is unlikely to have unduly influenced C_{resp} values for this species. It is unlikely that crushing had any significant effect on otolith-derived temperature.

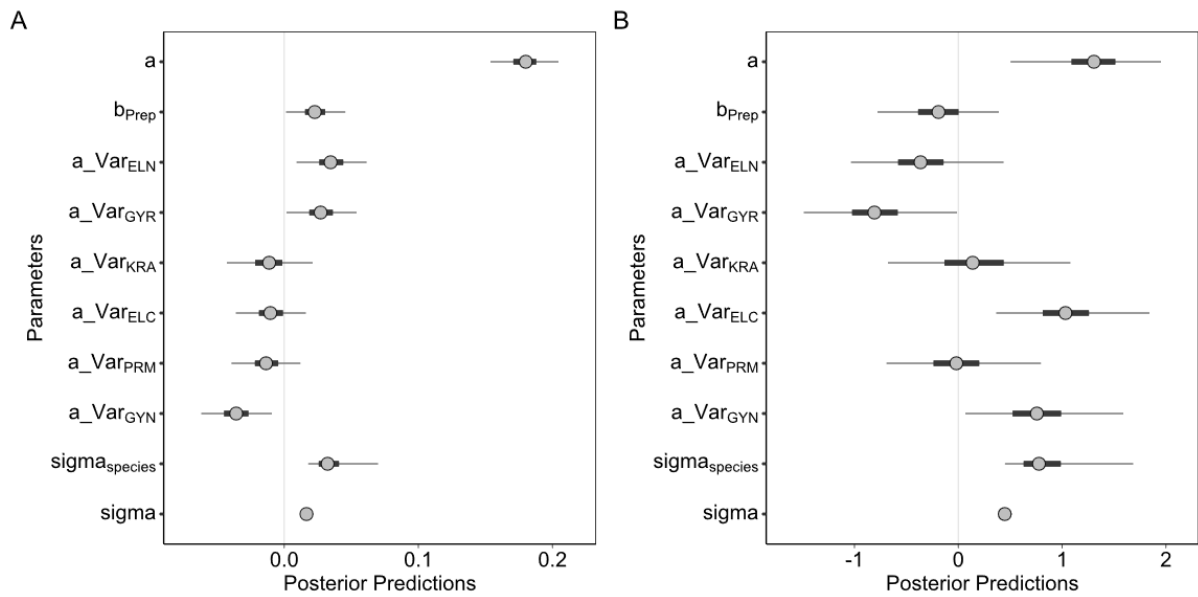


Figure S7: Posterior predictions for (A) equation S1 ($C_{resp} = a + b_{Prep} * Prep + a_Var_{Species}$) and (B) equation S2 ($Temp = a + b_{Prep} * Prep + a_Var_{Species}$). a is the intercept; b_{Prep} is the effect of crushing the otolith on C_{resp} and a_Var represents the variable intercept for each species; ELN = *Electrona antarctica*, GYR = *Gymnoscopelus braueri*, KRA = *Krefflichthys anderssoni*, ELC = *Electrona carlsbergi*, PRM = *Protomyctophum bolini* and GYN = *Gymnoscopelus nicholsi*. σ indicates overall residual error, and $\sigma_{Species}$ is residual error of the species variable intercept. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals, while thin lines show the 95% posterior intervals. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

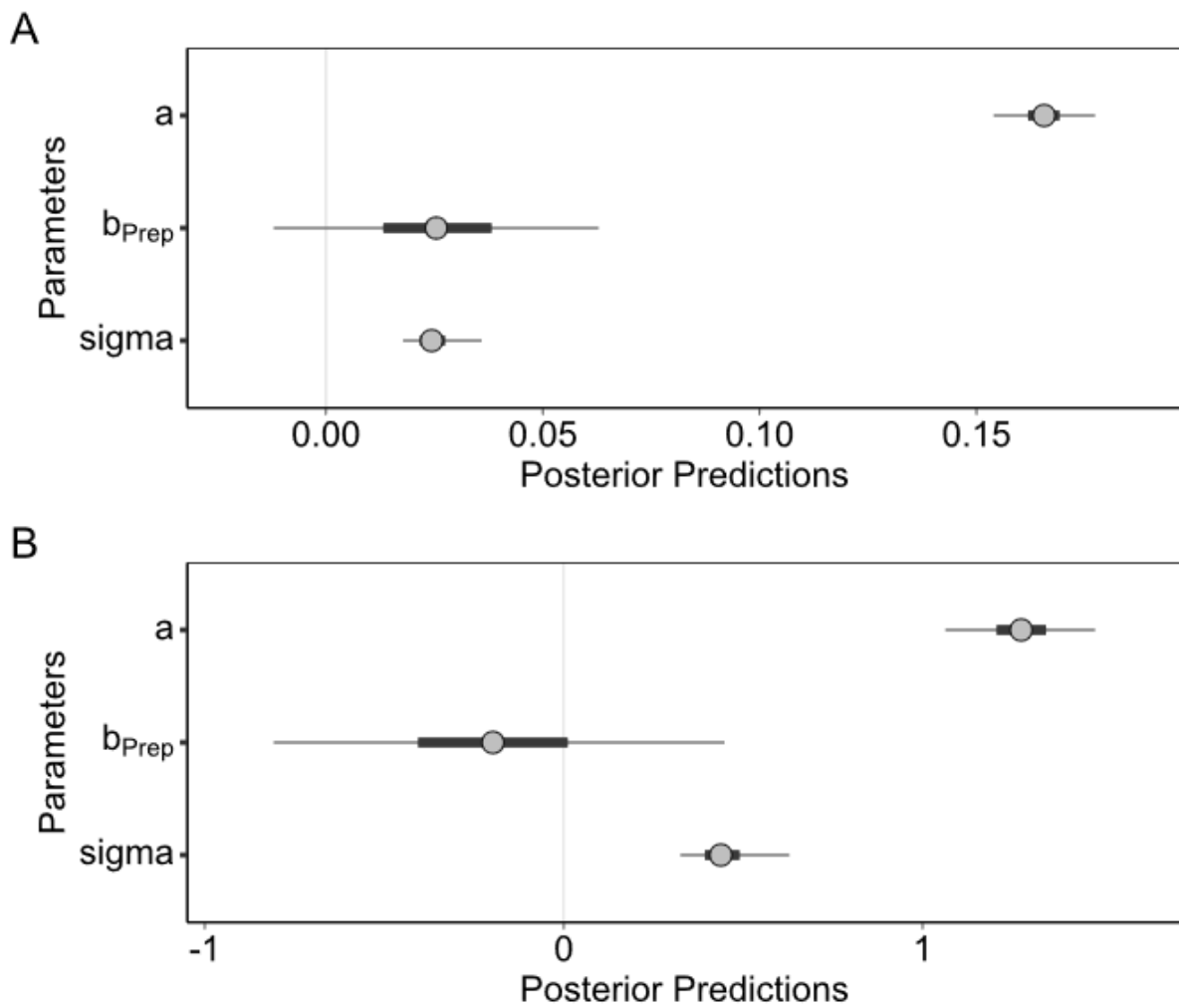


Figure S8: Posterior predictions for (A) equation S3 ($C_{resp} = a + b_{Prep} * Prep$), and (B) equation S4 ($Temp = a + b_{Prep} * Prep$), within *Protomyctophum bolini*. a is the intercept and b_{Prep} is the effect of crushing the otolith on C_{resp} and σ is the residual error. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals and thin lines show the 95% posterior intervals. Sigma indicates error. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

3 Effect life stage on C_{resp} values

To test for possible effects of different life stages on C_{resp} values, we divided individual standard length (SL, mm) by maximum SL for that species and used this to model C_{resp} :

$$C_{\text{resp}} = a + b_{R_{\text{SL}}} \times R_{\text{SL}} + a_{\text{Var}}_{\text{Species}} \quad (\text{S5})$$

Where R_{SL} is the ratio of SL to maximum SL and $b_{R_{\text{SL}}}$ is the effect of that ratio on C_{resp} . As with other models, we ran this in R version 4.0.5 (R Core Team 2021) with RStan version 2.21.2 (Stan Development Team 2020) using the rethinking package version 2.01 (McElreath 2020). We ran a single chain of 10000 iterations, 5000 warmup and a thinning parameter of one. Only milled otoliths were included in this model, as these samples best matched recorded SL for each individual. Although there appears to be a negative correlation between SL ratio and C_{resp} values (Figure S9), which supports the use of C_{resp} as a metabolic proxy, we found that there was no effect of this ratio on C_{resp} when accounting for replication within species (Figure S10).

We ran the same model within each species, without the random effect of species:

$$C_{\text{resp}} = a + b_{R_{\text{SL}}} \times R_{\text{SL}} \quad (\text{S6})$$

We found that there was no significant correlation between life stage and C_{resp} values within any of the species (Figure S11).

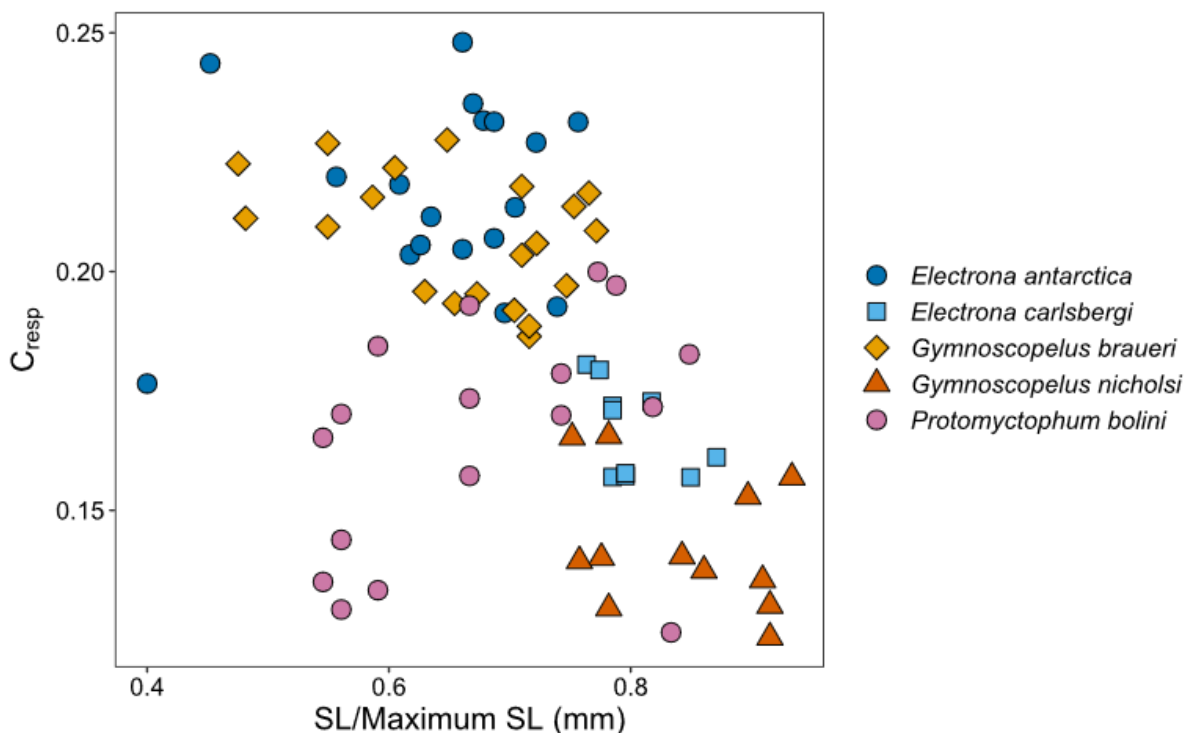


Figure S9: C_{resp} values plotted against ratio of standard length (SL, mm) of the individual to maximum SL for that species, for individuals of five myctophid species.

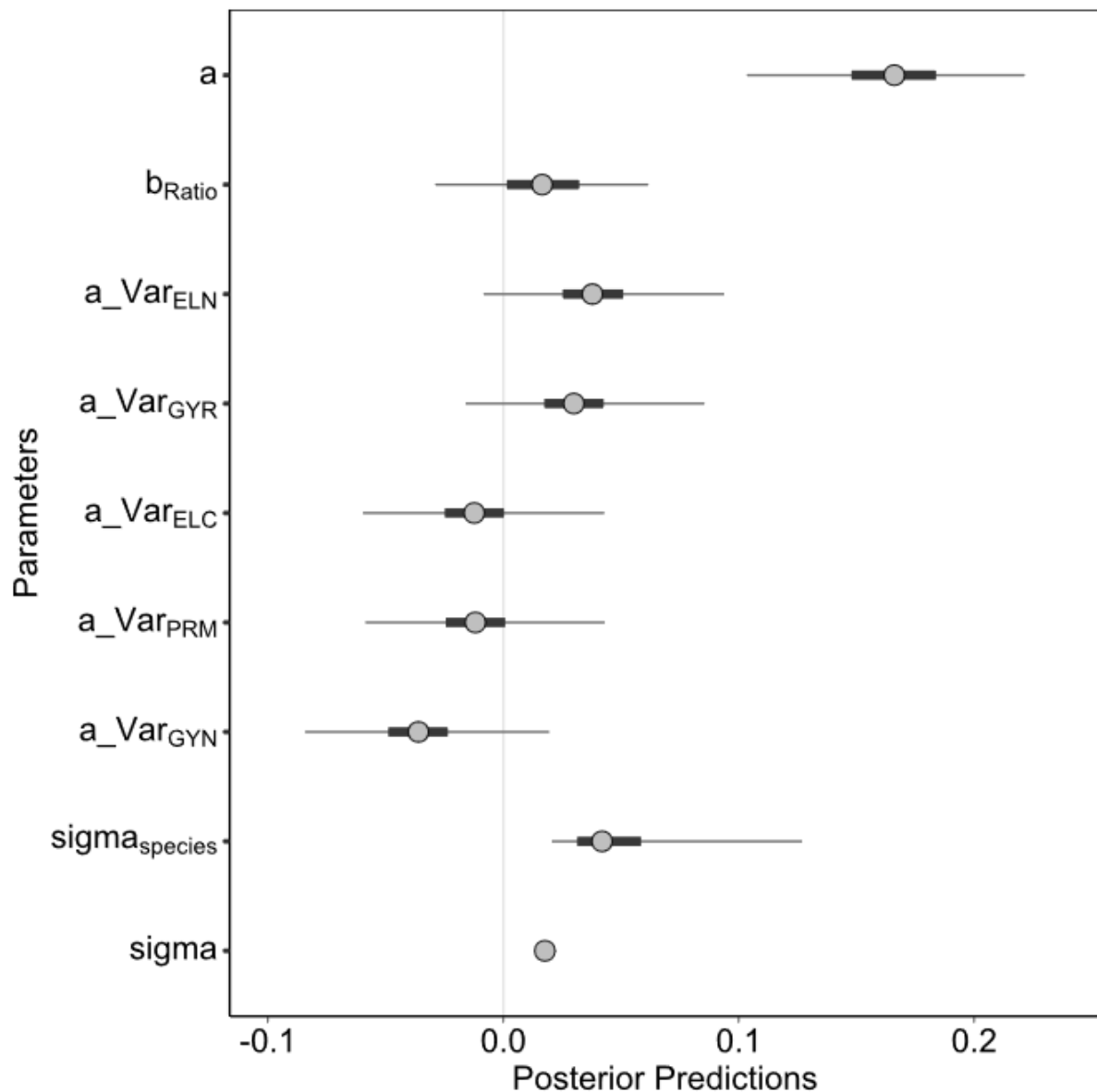


Figure S10: Posterior predictions for equation S5 ($C_{resp} = a + b_{R_SL} * R_{SL} + a_Var_{Species}$). a is the intercept; b_{R_SL} is the effect of the ratio of standard length (mm) to species maximum length on C_{resp} values, and a_Var represents the variable intercept for each species; ELN = *Electrona antarctica*, GYR = *Gymnoscopelus braueri*, KRA = *Krefflichthys anderssoni*, ELC = *Electrona carlsbergi*, PRM = *Protomyctophum bolini* and GYN = *Gymnoscopelus nicholsi*. σ indicates overall residual error, and $\sigma_{Species}$ is residual error of the species variable intercept. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals, while thin lines show the 95% posterior intervals. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

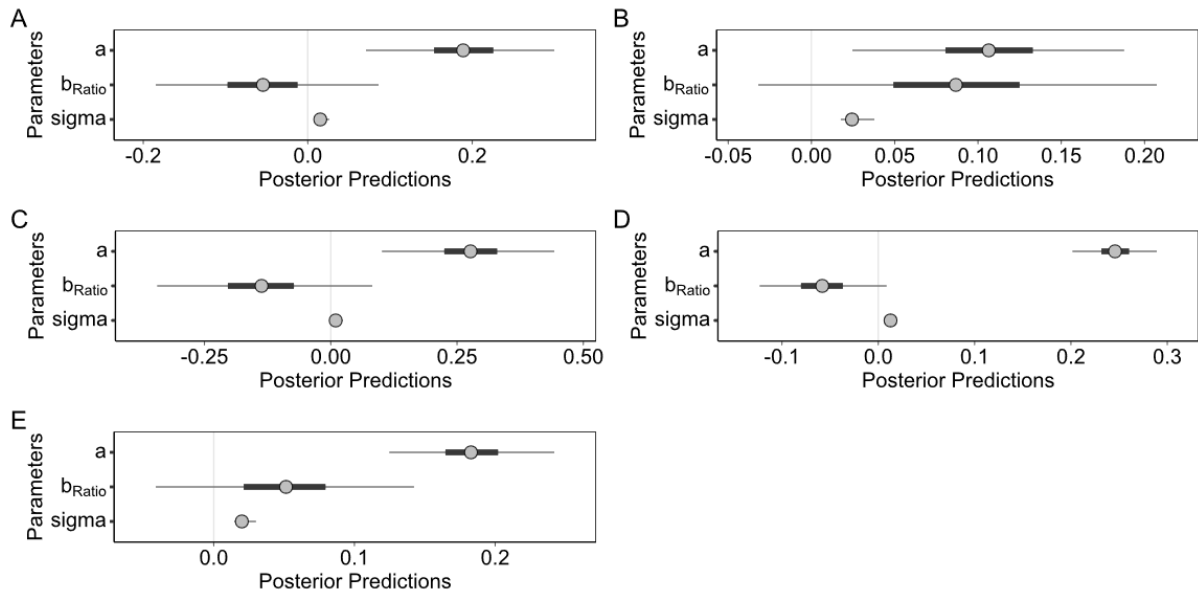


Figure S11: Posterior predictions for equation S6 ($C_{resp} = a + b_{R_SL} * R_{SL}$) within species (A = *Gymnoscopelus nicholsi*, B = *Protomyctophum bolini*, C = *Electrona carlsbergi*, D = *Gymnoscopelus braueri*, E = *Electrona antarctica*). a is the intercept and b_{R_SL} is the effect of the ratio of standard length (mm) to species maximum length on C_{resp} . Thin lines show the 95% highest density posterior intervals. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

4 Further investigations within species

We did the following analyses to aid in our understanding of the causes of intraspecific variation within certain species. As with other models, we ran this in R version 4.0.5 (R Core Team 2021) with RStan version 2.21.2 (Stan Development Team 2020) using the rethinking package version 2.01 (McElreath 2020). We ran a single chain of 10000 iterations, 5000 warmup and a thinning parameter of one.

4.1 Effect of year of capture within species

We used the following model investigate whether year of capture had a significant effect on C_{resp} within each species:

$$C_{\text{resp}} = a + b_{\text{Year}} \times \text{Year} \quad (\text{S7})$$

Where b_{Year} is the effect of year of capture on C_{resp} . Year of capture was assigned to a dummy variable (1998 = -1, 2008 = 0 and 2016 = 1). Within most species, year of capture had no significant effect on C_{resp} . *Protomyctophum bolini* was the exception (Figure S12), wherein individuals captured in 2016 had a higher mean C_{resp} than those captured in 2008 ($b_{\text{Year}} = 0.028 \pm 0.01$). This led to further investigations within *P. bolini* (see below).

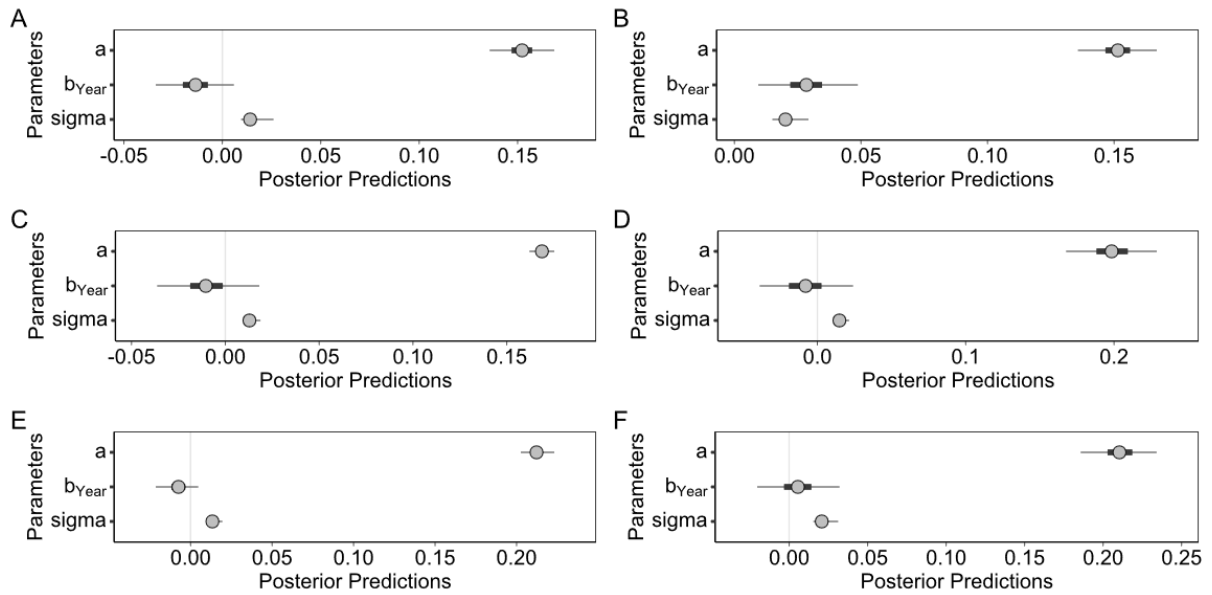


Figure S12: Posterior predictions for equation S7 ($C_{\text{resp}} = a + b_{\text{Year}} * \text{Year}$) within species (A = *Gymnoscopelus nicholsi*, B = *Protomyctophum bolini*, C = *Electrona carlsbergi*, D = *Krefflichthys anderssoni*, E = *Gymnoscopelus braueri*, F = *Electrona antarctica*). a is the intercept, b_{Year} is the effect of year of capture on C_{resp} values within that species, and σ is residual error. Circles are the mean of the posterior predictions. Thin lines show the 95% highest density posterior intervals. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

4.2 Further investigations within *Protomyctophum bolini*

To test whether latitude had a significant effect on C_{resp} within *P. bolini*, we modelled C_{resp} as a function of latitude of capture:

$$C_{\text{resp}} = a + b_{\text{Lat}} \times \text{Lat} \quad (\text{S8})$$

Where Lat is the latitude of capture in decimal degrees and b_{Lat} is the effect latitude of capture on C_{resp} values. (Stan Development Team 2020; McElreath 2020). *P. bolini* individuals captured further north did have lower C_{resp} values than those captured further south, as indicated by b_{Lat} 95% HDPIs not overlapping zero (Figure S13).

To investigate whether this phenomenon was specific to *P. bolini*, we ran the above model across all species, but also included a variable intercept of species ($a_{\text{VarSpecies}}$):

$$C_{\text{resp}} = a + b_{\text{Lat}} \times \text{Lat} + a_{\text{VarSpecies}} \quad (\text{S9})$$

When investigating all species together, there was no effect of latitude on C_{resp} values (Figure S14).

To investigate whether diet could be the driver of this variation, we modified equation S8 to test for an effect of latitude on $\delta^{13}\text{C}$ of muscle ($\delta^{13}\text{C}_{\text{musc}}$), our proxy for $\delta^{13}\text{C}$ of diet.

$$\delta^{13}\text{C}_{\text{musc}} = a + b_{\text{Lat}} \times \text{Lat} \quad (\text{S10})$$

We found no significant effect of latitude on $\delta^{13}\text{C}_{\text{musc}}$ (Figure S15).

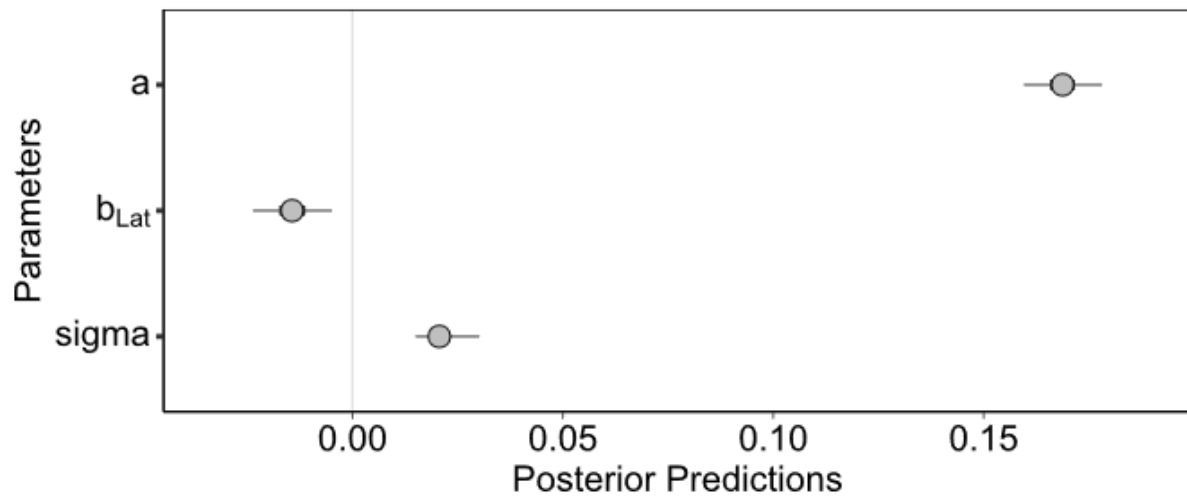


Figure S13: Posterior predictions for equation S8 ($C_{\text{resp}} = a + b_{\text{Lat}} * \text{Lat}$) within *Protomyctophum bolini*. a is the intercept and b_{Lat} is the effect of latitude of capture on C_{resp} and σ is the residual error. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals and thin lines show the 95% posterior intervals. σ indicates error. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

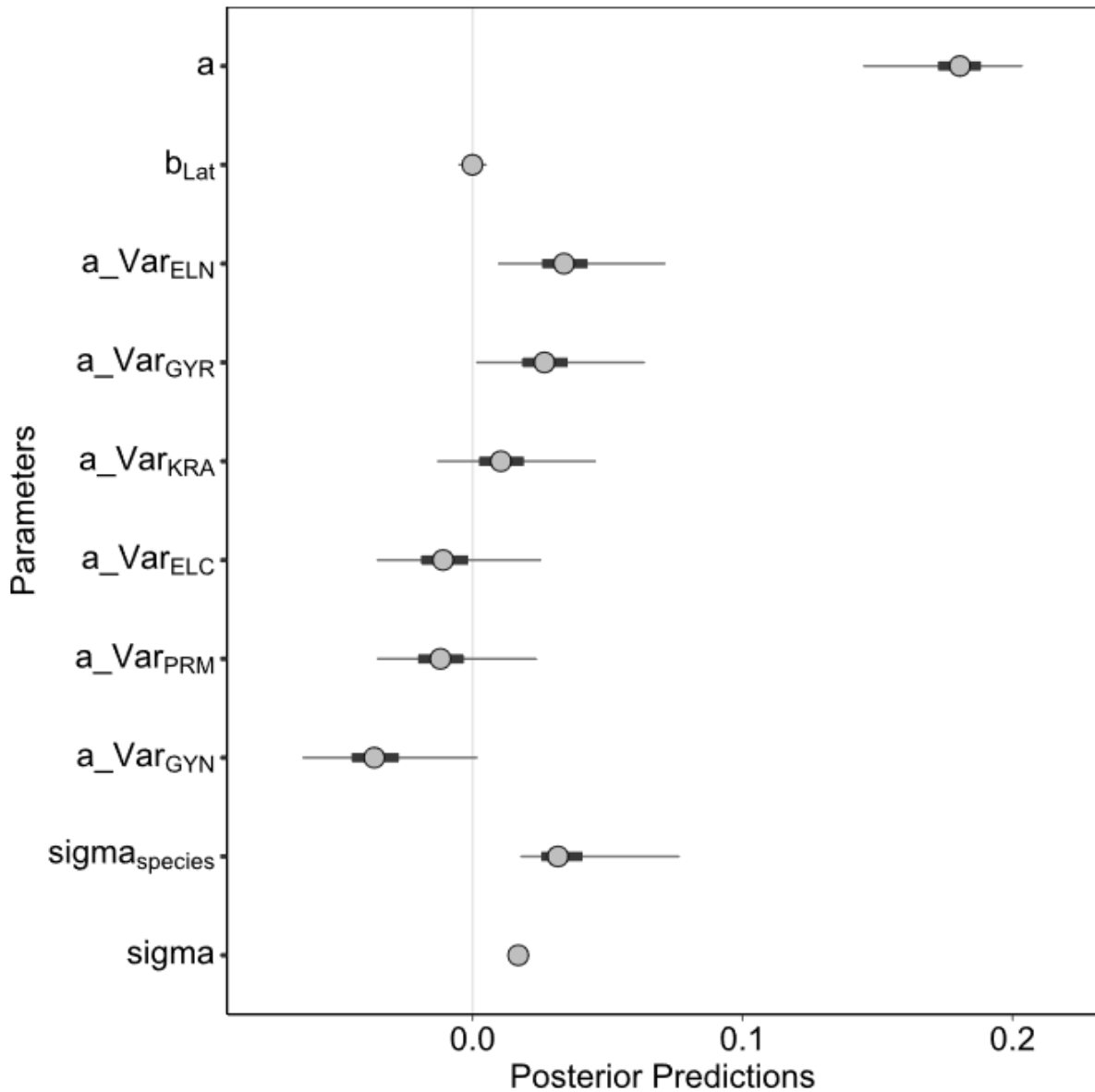


Figure S14: Posterior predictions for equation S9 ($C_{resp} = a + b_{Lat} * Lat + a_{Var_{Species}}$). a is the intercept; b_{Lat} is the effect of latitude on C_{resp} values, and a_{Var} represents the variable intercept for each species; ELN = *Electrona antarctica*, GYR = *Gymnoscopelus braueri*, KRA = *Krefftichthys anderssoni*, ELC = *Electrona carlsbergi*, PRM = *Protomyctophum bolini* and GYN = *Gymnoscopelus nicholsi*. σ indicates overall residual error, and $\sigma_{Species}$ is residual error of the species variable intercept. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals, while thin lines show the 95% posterior intervals. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

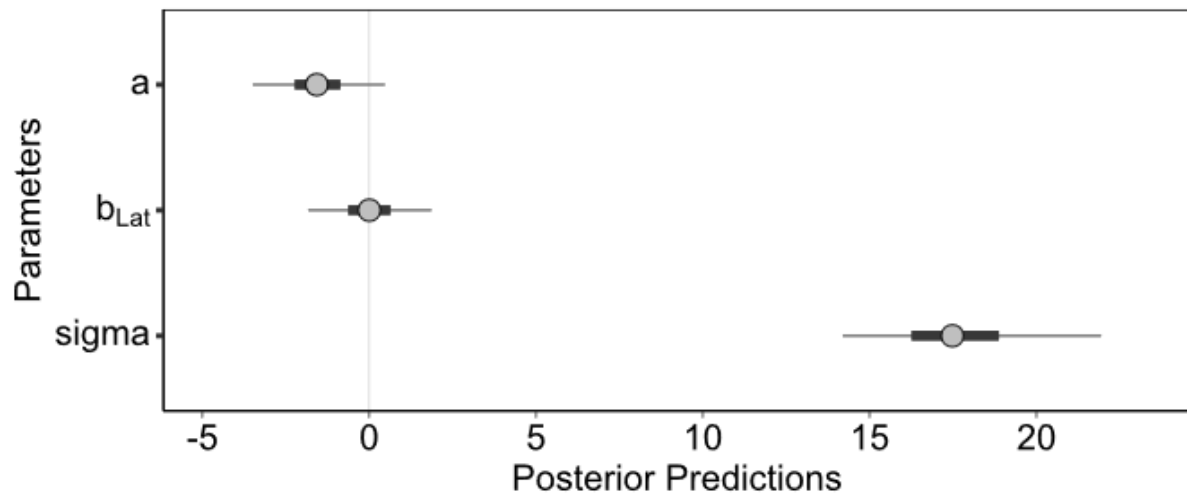


Figure S15: Posterior predictions for equation S10 ($\delta^{13}\text{C}_{\text{musc}} = a + b_{\text{Lat}} * \text{Lat}$) within *Protomyctophum bolini*. a is the intercept and b_{Lat} is the effect of latitude of capture on C_{resp} and sigma is the residual error. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals and thin lines show the 95% posterior intervals. Sigma indicates error. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

4.3 Further investigations within *Gymnoscopelus nicholsi*

We tested whether there was a significant difference between C_{resp} values of *G. nicholsi* individuals caught on the South Orkneys shelf breaks and those caught elsewhere. We did this by modelling C_{resp} as a function of location:

$$C_{\text{resp}} = a + b_{\text{Location}} \times \text{Location} \quad (\text{S11})$$

Where Location is the location of capture assigned to a dummy variable (South Orkneys = 1, elsewhere = 0) and b_{Location} is the effect of location being South Orkneys on C_{resp} values. There was no significant difference between C_{resp} values of *G. nicholsi* individuals captured at the South Orkneys compared to those captured elsewhere (Figure S16).

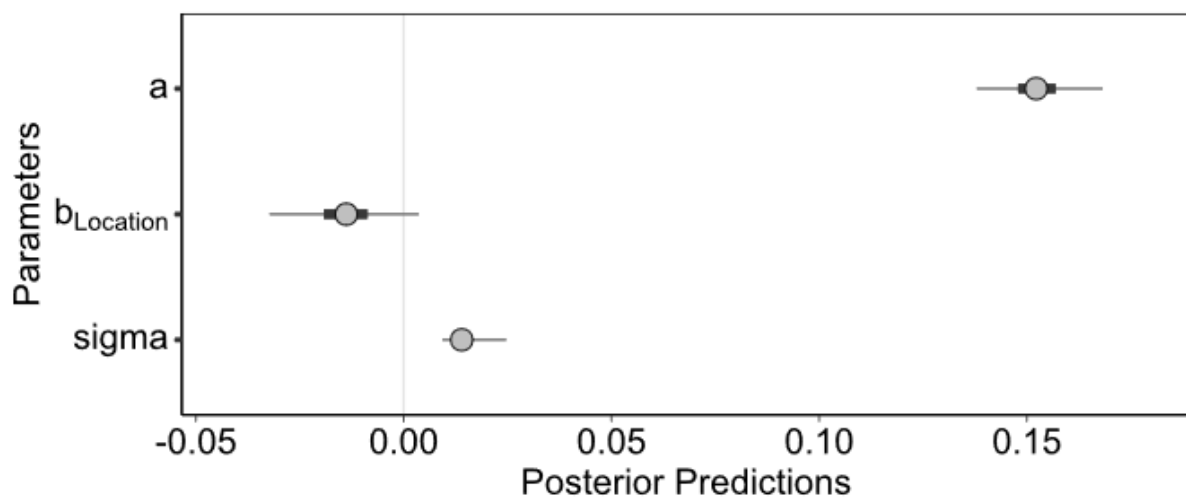


Figure S16: Posterior predictions for equation S11 ($C_{\text{resp}} = a + b_{\text{Location}} * \text{Location}$) within *Gymnoscopelus nicholsi*. a is the intercept and b_{Location} is the effect of catch location being the South Orkneys on C_{resp} and σ is the residual error. Circles indicate the mean of the posterior predictions. Thick lines show the 50% posterior intervals and thin lines show the 95% posterior intervals. σ indicates error. Results are considered statistically significant if the 95% highest density posterior intervals do not overlap with zero.

5 Age estimations of study individuals from length

For Table 2, we estimated the age (t) of each individual using a rearranged von Bertalanffy growth function:

$$t = \frac{1}{K} \log_e \frac{L_\infty}{L_\infty - L_t} + t_0 \quad (\text{S12})$$

Where K is the growth coefficient, L_t is standard length (mm) L_∞ is asymptotic standard length and t_0 is length at age 0. Table S1 gives the parameters used and their sources. Unfortunately there are no reliable growth parameters for *Protomyctophum bolini*.

Table S1: Growth function parameters for studied species. SL is standard length, L_∞ is asymptotic standard length, K is the growth coefficient and t_0 is length at age 0. ELN = *Electrona antarctica*, GYR = *Gymnoscopelus braueri*, KRA = *Krefflichthys anderssoni*, ELC = *Electrona carlsbergi* and GYN = *Gymnoscopelus nicholsi*.

Species	SL Units	L_∞	K	t_0	Source
ELN	cm	11.3	0.21	0.7	Linkowski (1987)
GYR	mm	133.22	0.29	-0.21	Saunders et al. (2020)
KRA	mm	68.6	0.71	-0.49	Saunders et al. (2020)
ELC	cm	9.7	0.55	-0.6	Linkowski (1987)
GYN	mm	163.8	0.41	0.081	Linkowski (1985)

References

- Linkowski T (1985) Population biology of the myctophid fish *Gymnoscopelus nicholsi* (Gillbert, 1911) from the western South Atlantic. *J Fish Biol* 27:683-698
- Linkowski T (1987) Age and growth of four species of *Electrona* (Teleostei, Myctophidae) Proceedings of the 5th Congress of European Ichthyologists. Swedish Museum of Natural History Stockholm, p 435-442
- McElreath R (2020) *rethinking: Statistical Rethinking book package*
- R Core Team (2021) *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria
- Saunders RA, Lourenço S, Vieira RP, Collins MA, Assis CA, Xavier JC (2020) Age and growth of Brauer's lanternfish *Gymnoscopelus braueri* and rhombic lanternfish *Krefflichthys anderssoni* (Family Myctophidae) in the Scotia Sea, Southern Ocean. *J Fish Biol* 96:364-377
- Stan Development Team (2020) *RStan: The R interface to Stan*