



Estimating fossil biomass from skeletal mass in marine invertebrates

CAITLIN A. MEADOWS

LETHAIA



Meadows, C.A. 2019: Estimating fossil biomass from skeletal mass in marine invertebrates. *Lethaia*, Vol. 52, pp. 323–334.

Palaeoecology uses the numerical abundance and the occurrence of species to evaluate the dynamics of past communities, but biomass – the quantity of soft tissue – is the critical currency needed to capture the flow and role of nutrients in modern ecosystems. Acquiring biomass data from fossil assemblages has, however, remained challenging, thus limiting the analysis of net secondary production in palaeocommunities. Prior models relate shell size or shell biovolume to fossil biomass. These models neglect shell fragments and, moreover, use units of biovolume (cm^3) that are not directly related to those of biomass (g), making the models difficult to tune and the coefficients highly specific. To remedy these shortcomings, I evaluate skeletal mass as a means of estimating the soft tissue biomass of fossil taxa, using ratios among biomass, skeletal mass and the total wet mass of living representatives of extant species, so that skeletal mass alone can be used to estimate grams of organic biomass. Data on total wet mass, organic carbon mass, and shell mass were acquired from more than 80 live-collected individuals from eight families in three major, shelly macrobenthic groups (Mollusca, Brachiopoda, Arthropoda) and supplemented with counterpart data from the literature to increase taxonomic breadth. This new shell-mass model provides more accurate and precise biomass estimates than models based on the linear dimensions of shells, expanding our ability to examine the interplay between organisms and their environments. □ Biomass, carbonate mass, fossil, macrofauna, organic carbon, skeletal mass.

Caitlin A. Meadows [meadowsc@uchicago.edu], The Department of the Geophysical Sciences, The University of Chicago, 5734 S Ellis Ave Chicago, IL 60637, USA; manuscript received on 2/01/2018; manuscript accepted on 6/10/2018.

In both modern and ancient ecosystems, biomass is an index of how organic resources are consumed and converted into living tissues and then ultimately recycled (Staff *et al.* 1985; Bambach 1993; Powell *et al.* 2001; Brown *et al.* 2004; Payne & Finnegan 2006). In modern communities, biomass usually reflects the soft tissues of all live-collected animals, both shelled and non-shelled, and so this currency presents many challenges to the palaeoecologist confronted with only shelly remains. The study by Staff *et al.* (1985), using macroinvertebrate communities in two Texas lagoons, found that the total preserved fossil assemblage (mostly molluscs) resembled the whole living community and the preservable subset of that community more closely when compared using units of 'biomass' than using numerical abundance. Biomass data thus appear to show higher fidelity to live-assemblage counterparts, in addition to revealing information on macrobenthic productivity not provided by abundance data alone. Biomass data provide an essential complement to palaeoecological insights already gained from abundance and occurrence data regarding population size, successful reproductive strategies and habitat occupancy (Staff *et al.* 1985; Bambach 1993; Forcino *et al.* 2010, 2015; Allmon & Martin 2014). Biomass data can also be used as the

basis of more complex models to estimate the net secondary production and trophic demands and structure of fossil assemblages (e.g. Powell *et al.* 2001; Finnegan 2013).

In modern communities, biomass is often measured by either incineration or wet oxidation of the soft tissues of live-collected animals and is usually expressed as either the (dry) mass of those tissues (ash-free dry mass; AFDM) or the mass of only the organic carbon (C_{org}) in those tissues (Table 1; e.g. Brey *et al.* 1988). By contrast, the biomass of a fossil assemblage must be extrapolated from the preserved skeletal elements. These approximations use linear dimensions of complete (unbroken) individual fossils to approximate the original 'biovolume' of the animal, from which a mass of organic soft tissue is estimated based on species-specific regression coefficients, calibrated using live representatives (Powell & Stanton 1985; Staff *et al.* 1985). The need for nearly complete fossils limits the number of individuals in a fossil assemblage that can be used to estimate biomass this way.

To alleviate these limitations, the biomass of fossils has also been estimated either by using the skeletal (usually carbonate) mass as a proxy for soft tissue-based biomass (Waselkov 1987; Forcino *et al.* 2010) or by extrapolating the edible 'meat mass' from the

Table 1. Common measures of biomass, with abbreviations of variables used in this paper in the first column. Definition is the agreed upon definition of the value from the following sources (1) Brey *et al.* (1988), (2) Palmerini & Bianchi (1994), (3) Brey *et al.* (2010), (4) Palmer (1982), (5) Forcino *et al.* (2010) and (6) Steimle & Terranova (1985). All masses measured in grammes.

Variable name	Abbreviations in the literature	Synonyms	Common name	Definition in published source
W	WM	Tot _{wet}	Total wet mass	Total wet body mass including soft tissue, skeletal mass and with or without water held in body cavities (1, 2, 3)
	SFWM	WM	Shell-free wet mass	Wet mass of only the soft tissue dissected from the shell; sometimes called WM if shells are easily removed from body (1, 2, 3)
	DM	DM + Shell	Dry mass	The combined shell and tissue mass of an organism after drying; can either include or exclude shell and so must be clearly defined (2, 3)
	SFDM	DM	Shell-free dry mass	Only the soft tissue dried in furnace until constant mass (1, 2, 3)
	Ash		Ash	The residue left from incinerating the dry soft tissue, burning off all organic material (2)
B	AFDM	B _{org}	Ash-free dry mass (organic biomass)	The burned-off mass from tissue incineration, represents organic material biomass (1, 2, 3)
C	C _{org}	g carbon	Organic carbon biomass	Grams carbon burned off of a tissue sample in a bomb calorimeter or wet oxidation experiment (1, 2)
S	SM	Submerged Mass	Shell mass	The mass of only the dissected shell as weighed separately from the soft tissue; can be found from or approximated from submerged mass (4)
x	FM	Calcified biomass	Fossil mass	The mass of the preserved shell material in a fossil assemblage (5)
	Water _{loss}	H ₂ O content	Water loss	Water lost from drying and/or doing dissection (6)

skeletal mass in a shell midden (Lyman 2008). In these types of shell-mass approaches, fossils must be preserved in their original mineralogy and porosity (shell density), a condition common in dead-shell assemblages on modern seafloors and in many unlithified sediments. Focusing on skeletal mass also facilitates analysis of the mass of calcium carbonate produced in an assemblage or ecosystem, which is a valuable metric of the inorganic carbon cycle (Davoult *et al.* 2009; Lejart *et al.* 2012; Waldbusser *et al.* 2013).

Here, combining new measurements of shell mass, soft tissue mass and organic carbon content in living congeners with a correction for fragmented shells in the target fossil assemblage, I demonstrate a new model that can estimate biomass for fossil assemblages more directly than prior models and can accommodate both fragmented and colonial fossil individuals.

Approach

Measuring living biomass

Biomass is most commonly quantified either as AFDM or as organic carbon (C_{org}), which approximate the acquisition of organic *material* or organic *carbon* into the assemblage, respectively (Brey *et al.* 1988, 2010; Brey 2001). Other commonly reported biomass metrics are total wet mass

(WM); shell-free mass, also known as soft tissue wet mass (SFWM); and shell-free dry mass (SFDM) (Table 1; Fig. 1; Brey *et al.* 1988). The biomasses enumerated here do not quantify energetic production (calories, kJ) but instead focus on organic production. Directly measuring organic biomass and organic carbon biomass require dissecting, drying and incinerating every surveyed individual (Fig. 1; Palmerini & Bianchi 1994), which is both time-consuming and destructive. To alleviate these costs, biologists take a simplified, indirect approach, applying coefficients developed using exemplar sets of living individuals. The organic biomass (B, Eqn 1a) or organic carbon (C, Eqn 1b) of a given taxon in a sample is approximated as the proportion of the measured total wet mass (W) of animals that is organic matter (α_b) or organic carbon (α_c ; Table 2).

$$B = \alpha_b W \quad (1a)$$

$$C = \alpha_c W \quad (1b)$$

Ratio of biomass to total wet mass (α). – Since they were first applied to invertebrate fisheries by Thorson (1957), biomass coefficients related to total wet mass have been gathered from many taxa, accumulated in databases and tested for accuracy and efficiency through meta-analysis (Steimle & Terranova 1985; Rumohr *et al.* 1987; Palmerini & Bianchi 1994; Ricciardi & Bourget 1998; Brey *et al.*

2010). Consequently, these masses and their proportions (α_b and α_c) have become standard metrics.

These studies reveal that the majority of variation in α_b and α_c values owes to the effect of the water content of the animal, that is, the liquid in body cavities and the liquid in the tissues, which varies strongly among groups. Water in mantle cavities ranges between 10% and 40% of total wet mass, for example, 40% in Mytilidae, which have large mantle cavities, but only 10–15% in Tellinidae calculated from values reported in Rumohr *et al.* (1987). The remaining water is lost in the drying process and accounts for 25–43% of the total wet mass of bivalves and 17–38% in gastropods (Rumohr *et al.* 1987). Some families exhibit sizeable interspecific variation in water content and therefore broader ranges in α_b . For example, for the bivalve family Cardiidae, total water mass ranges between 45% and 68% of the total wet mass resulting in a broad range of α_b among species (0.05–0.08), whereas Astartidae in the same study has a much lower and less variable range of total water mass (23–30% of total wet mass) and a narrow α_b range among species (0.05–0.062; Rumohr *et al.* 1987).

Water content and thus α_b and α_c can also vary naturally among individuals within species and populations as well as over time for single individuals as a function of the season, animal life stage and health of the individual. These variations are typically minimal, for example, α_b ranges among individuals and between seasons from 0.016 to 0.057 in the bivalve *Corbicula manilensis* per Aldridge & McMahon (1978). In contrast, variation in α_b among species

Table 2. Definitions and equations of the coefficients used in this study.

Variable name	Equation	Definition
α_b, α_c	$\alpha_b = \frac{B}{W},$ $\alpha_c = \frac{C}{W}$	The ratio between organic biomass (B) or organic carbon (C) to an individual's total wet mass (W)
β	$\beta = \frac{S}{W}$	The ratio between an individual's shell mass (S) and its total wet mass (W)
γ	$\gamma = \frac{x}{S}$	The ratio between the preserved fossil (x) and an ideal complete shell or skeletal element (S)

within a family can be quite considerable, for example, Buccinidae range between 0.048 in Steimle & Terranova (1985) and 0.177 in Stoker (1978).

Other sources of variation in coefficient values can arise from interlaboratory differences in sample processing, such as decanting mantle fluids before weighing, mechanically or chemically separating shell from flesh or incinerating only tissue or the entire organism. Palmerini & Bianchi (1994) explored these sources of variation in *Mytilus galloprovincialis*. They found that although α_b can vary up to 48%, Equation (1) continually returned reliable estimates of AFDM regardless of the exact laboratory procedures used (table 4 in Palmerini & Bianchi 1994). Thus, the variability introduced by interlaboratory differences in sample processing is small compared to the variation in population sizes over a season-long or multi-year biological survey. Many modern studies evaluating an entire ecosystem typically make use of family-level (e.g. Grebmeier *et al.* 1988) or class level (Ricciardi & Bourget 1998), with species-level values used only when needed for a key taxon or when available (Brey *et al.* 2010).

Estimating fossil biomass

Building on this established biological method, I use skeletal mass to estimate the organic biomass represented by a preserved fossil individual, using equation 2. This model requires two mass conversion factors (α_b and β), each calibrated using live-collected individuals of the target species or a living congener. The model also requires an estimate of post-mortem damage (γ) applied to the mass of the fossil individual (x). The organic biomass of a fossil is

$$B = \frac{\alpha_b x}{\beta \gamma}, \quad (2)$$

α_b is, as described above, the ratio of soft tissue mass (B) to total wet mass (W), and β is the ratio between shell mass (S) and total wet mass (W), calibrated

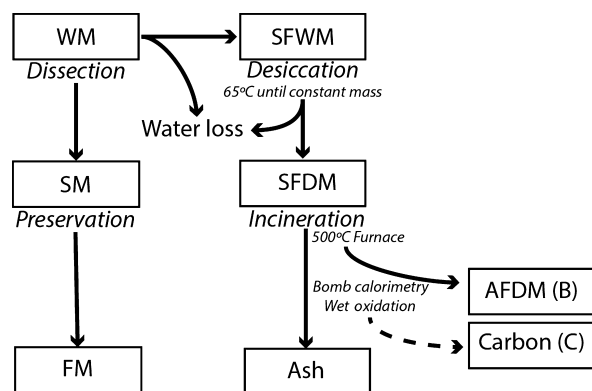


Fig. 1. Flowchart for acquiring biomass metrics described in Table 1, based mostly on Palmerini & Bianchi (1994) and using abbreviations commonly found in the literature (Table 1). In order of increasing processing effort: WM = total wet mass (may or may not include 'water content' of body cavities), SM, shell mass; FM, fossil mass; SFWM, shell-free wet mass; SFDM, shell-free dry mass; AFDM (B), ash-free dry mass, that is organic material biomass only; Carbon (C), organic carbon biomass; Ash, the ash produced from incineration.

with live-collected individuals (Table 2). Shifting to values acquired from the fossil material: x is the mass of the measured fossil shell, which may be complete or a fragment; γ is the proportion of the original shell preserved as a fossil, that is, the ratio of shell mass (x) to the total weight (S) of a complete, unbroken shell (Table 2). γ can be estimated by comparing the shell to the outline of a complete, undamaged shell of the same taxon (see below the ratio of measured fossil to undamaged shell (γ)). The coefficient α_b in Eqn 2 can replace by α_c to calculate the organic carbon biomass of a fossil (C) (Table 2).

Ratio of shell mass to total wet mass (β). – The coefficient β is the proportion of shell mass (S) to total wet mass (W) (Table 2). β is less commonly reported than other biomass metrics because shells are generally discarded after dissection (Fig. 1). However, the few published β values could be supplemented by calculating β from other more widely reported proportions, such as $1 - (W/(W + S))$ (Brey *et al.* 2010).

Sources of variability in β are similar to those in α and can be similarly quantified. Shell thickness varies naturally as a function of life stage, growth rate and ornamentation, all of which vary with species, latitude and ocean chemistry. Of these effects, the increase in relative shell mass with body size – shell thickening over an animal's lifespan, especially in molluscs – is most well-known (e.g. Aldridge & McMahon 1978; Stoker 1978) and accounts for ± 0.05 β values per family, for example, the bivalves Cyrenidae in Aldridge & McMahon (1978) and Tellinidae in Stoker (1978).

Again, as with α , β can be strongly affected by the water content of an animal and is subject to the same variations discussed in the previous section. β generally varies between 0.25 and 0.75, with strong variation among species, for example, 0.362–0.661 in Mytilidae in Ricciardi & Bourget (1998). Much of this variation is likely due to morphologic differences. Unfortunately, the magnitude and controls on variation in β are not well supported by multiple databases and meta-analyses, unlike variation in α .

Ratio of measured fossil to undamaged shell (γ). – Fossil mass is likely to differ from the shell mass measured by a biologist – specifically, to be smaller – because of the long post-mortem interval separating fossils from their soft bodies: residual shells will typically accrue damage from exposure to the depositional environment. γ represents a gross estimate of this post-mortem damage, here quantified as fragmentation. The original skeletal

mass can be reconstructed for a fossil using the relationship

$$S = \frac{x}{\gamma}, \quad (3)$$

where S is the original full shell mass, x is the measured fossil mass, and γ is an estimate of fossil completeness relative to an undamaged shell, using shell outline. For example, a complete bivalve with $\gamma = 1$ would be a still articulated and undamaged two-valved shell; an isolated but complete valve would have $\gamma = 0.5$; and half of a disarticulated valve would have $\gamma = 0.25$ (Table 2; Fig. 2).

Methods

Acquiring α and β

To obtain the necessary coefficients to estimate biomass using shell mass reliably, I compiled values of α_b , α_c and β reported in the literature for marine shelled macrobenthos, drawing mainly on the online databank of Brey *et al.* (2010) (see additional references in the caption of Fig. 3).

To increase the taxonomic scope and sample sizes per taxon, I also measured the biomass of live-collected specimens and calculated their α_b , α_c and β values. These taxa included molluscs (Bivalvia: *Geukensia demissa*, *Mytilus edulis*, *Mercenaria mercenaria*, *Venerupis philippinarum*; Gastropoda: *Nucella lamellosa*, *Nucella osterina*, Littorinidae), shelled barnacles (Balanidae) and articulate brachiopods (*Terebratulina* sp., *Terebratula* sp., *Laqueus* sp.). Five to 15 individuals per species were processed to acquire the linear dimensions of shells (mm) and the following masses: shell mass (S), total wet mass (W) and biomass as AFDM. Shells were manually dissected and the tissues dried in a furnace at 65°C until they reached a constant mass (~48 h, SFDM). The tissues were incinerated in a furnace at 500°C for 3 h to attain the ash mass of the tissue, and AFDM (Fig. 1). Organic matter in the shell itself (organic matrix) was not measured because it is usually minimal (typically less than 1% of the shell mass and commonly only tenths or hundredths of a per cent) and is quickly lost post-mortem (Aldridge & McMahon 1978; Cameron *et al.* 1979; Glover & Kidwell 1993). Mean family values were then generated from calculations of α_b and β for each individual.

To characterize a family, a weighted grand mean was calculated and displayed with the full range of values (Fig. 3), using all reported mean values from

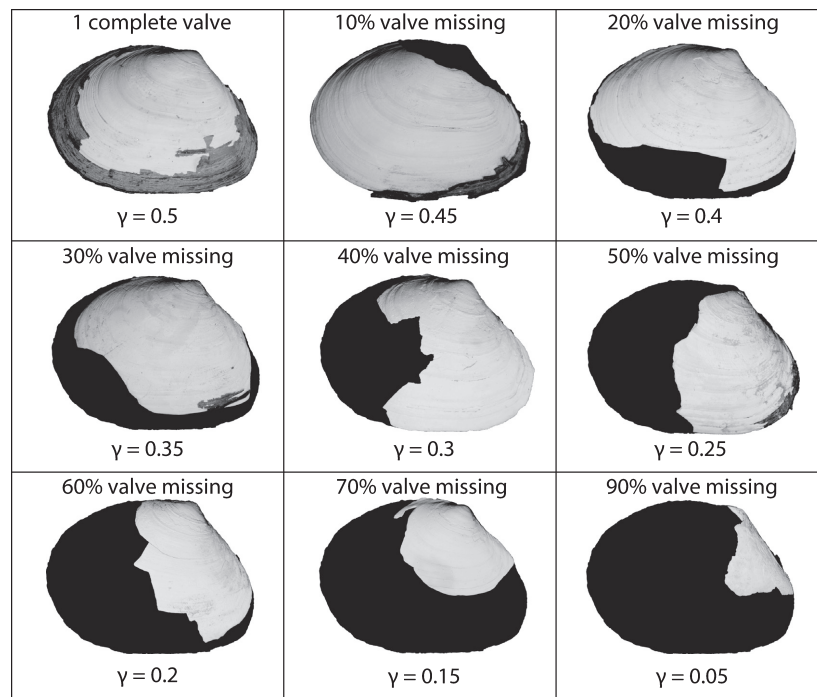


Fig. 2. Comparative chart for visually estimating gamma, the proportion of a fossil preserved relative to a full undamaged shell, using dead-collected specimens of the bivalve *Macoma calcarea* with natural patterns of breakage. Black shadows show the amount of material missing, as confirmed using polygon area in Image J. Exemplar shells are from multiple sites in the S Chukchi Sea, in 45 m water depths.

the literature (grey) plus the mean values newly produced here (white box whiskers), weighting each mean based on the study size (table of data available from author). Animal classes (bivalves, gastropods, etc.) were characterized by the median and the interquartile range (IQR) calculated from constituent families' means.

Relative accuracy of biomass models

To determine the comparative accuracy of biomass estimates, this new shell-mass model was compared to a linear-dimension (biovolume) biomass model. Biomass was calculated for each family both using equation 2 with mean family-level values of α_b and β (assuming $\gamma = 1$) and using a shell-dimension model (Eqn 4) with a mean family-level estimate of shell size

$$B = aL^b \quad (4)$$

where a and b are coefficients that are empirically fit from the measured data, and L is a linear dimension of the shell material, such as height (dorsal-ventral) or length (anterior-posterior).

Each calculated biomass, using either shell mass or linear-shell dimensions, was compared to that individual's organic biomass as measured by incineration

for every live-collected species. I also used (Palmer 1982) unique published data set for the length, total wet mass and shell mass of muricid gastropod individuals. In ideal circumstances, the shell-mass and linear-dimension methods of estimating biomass would perfectly coincide with measured biomass. The relative accuracy of these methods is determined by their correlation with measured organic biomass as expressed as r^2 .

Reliability of scoring the completeness of fossil specimens

To test the effect of γ on biomass estimates, I measured the shell height and shell mass of 295 individuals of the bivalve *Macoma* spp. collected from death assemblages in the N Bering and Chukchi Seas. Shell mass was reconstructed for each fossil individual using two models: (1) equation 3 based on fossil mass; and (2) a reconstruction based on fitting shell height to weight using only the complete specimens of *Macoma* ($\gamma \geq 0.95$) gathered in this analysis. To ensure that individuals were not double-counted in these tests, only fossil specimens preserving at least 50% of their hinge line were used; moreover, the linear dimension between umbo and ventral margin (bivalve height) had to be measurable. Shell height was used as the critical dimension rather than shell

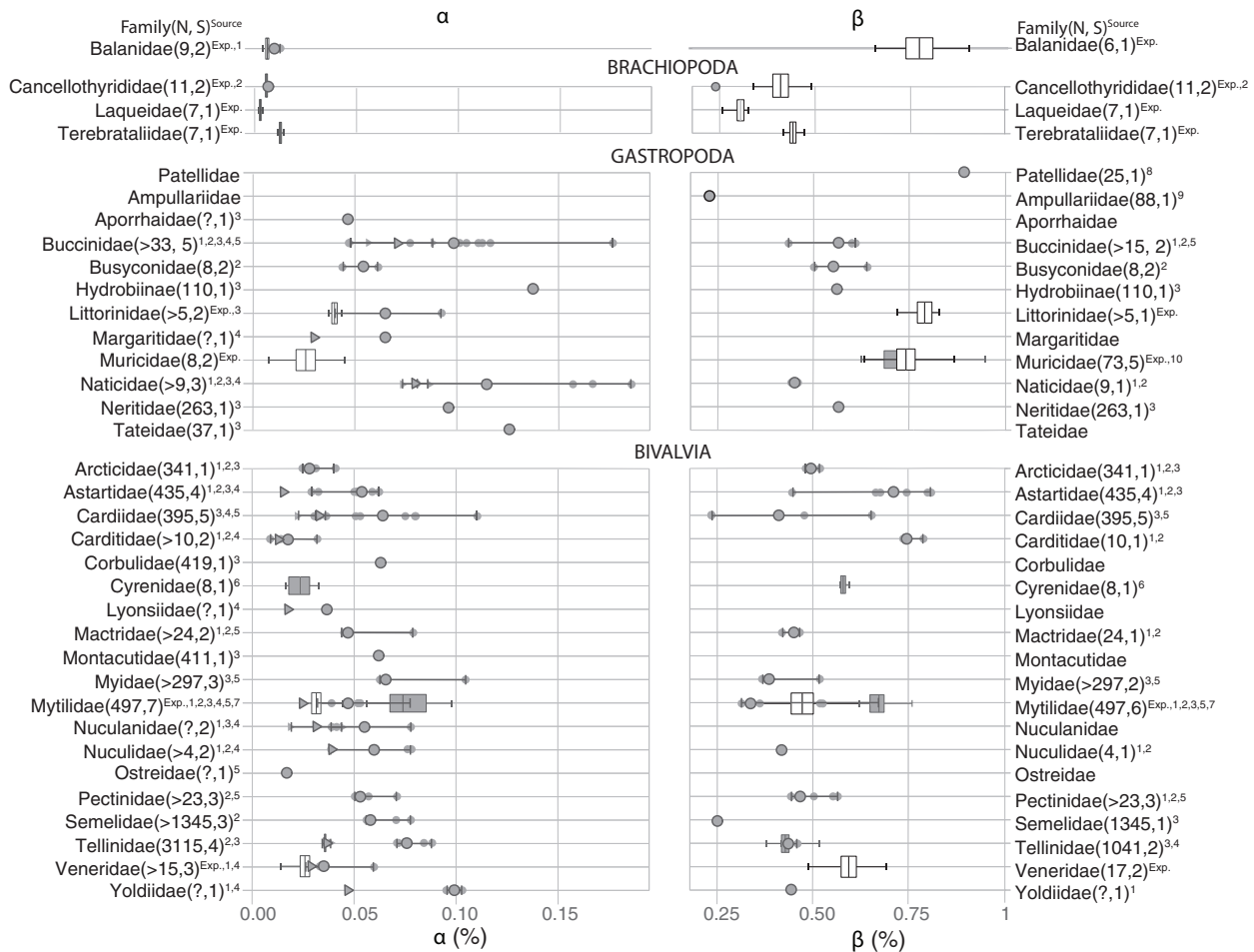


Fig. 3. Compilation of mean α and β values for shelly macro-invertebrates, either acquired from published reports or newly produced for this study, arranged alphabetically by family within class or phylum. Numbers after each family name denote the number of individuals (N), and species (S) used to produce the means. On the left, grey circles represent published α_b (B/W), and grey triangles represent published α_c (C/W). On the right, grey circles represent published β values (S/W). Box plots, show mean (dark line), standard deviations above and below (box), and the full range of values reported (whiskers). Grey box plots are published distributions of values as opposed to the solitary mean values. Box plots in α represent α_b values, except those in Tellinidae and Cyrenidae which represent α_c values. White box plots are data generated for this study. Finally, large grey and black outlined points are weighted means of each family. Weighted means were calculated weighted by the number of specimens used to calculate each mean. Whiskers from the large value span the entire distribution of reported values. Sources: (Exp.) original data generated here, (1) database of Brey *et al.* (2010), (2) Steimle & Terranova (1985), (3) Rumohr *et al.* (1987), (4) Stoker (1978), (5) Ricciardi & Bourget (1998), (6) Aldridge & McMahon (1978), (7) Palmerini & Bianchi (1994), (8) Cravo *et al.* (2004), (9) Guedes *et al.* (1981) and (10) Palmer (1982).

length because height is more often preserved among dead shells of *Macoma*.

The value of γ was estimated by a single operator using the comparative chart in Fig. 2 as a guide, and only the individuals with a measurable height were used in this analysis. Damage (shell thinning) from abrasion and corrosion was ignored in this study. These height-based estimates of shell mass, using the empirical relationship $S = 0.0001(H)^{3.0145}$, were treated as expected masses, and those estimated from the mass-based Eqn 3 were treated as the modelled shell mass. This approach allows the calculation of the chi-square error between the shell-height model and the shell-mass model presented here

(χ^2 error = $(\text{Obs} - \text{Exp})^2/\text{Exp}$). Residuals were also calculated by subtracting the shell-mass model result from the expected shell-height model result, to determine whether the shell-mass reconstructions under- or overestimated shell mass compared to the height model. In ideal circumstances, the modelled shell mass should slightly underestimate the true total shell mass owing to the likelihood of some post-mortem loss, producing a small chi-square error, a negative sum simple residual and a near-zero negative average simple residual.

To evaluate operator error in γ , two persons counted *Macoma* specimens from the death assemblages of two Van Veen samples, each yielding more than 125

individuals, using the same visual comparison guide for γ (Fig. 2). The maxima, minima, modes and interquartile ranges of reported γ were measured for each sample by each operator. A Wilcoxon signed-rank test, designed to test the similarity of paired non-normal datasets (McDonald 2009), was performed to test the reliability between observers statistically.

Results

Variation in α and β

Carbon biomass as a proportion of wet mass (α_c) is consistently low with a median of 0.028 and IQR of 0.019–0.037, among the 19 families of bivalves tested (Fig. 3). α_b is slightly higher and more variable with a median of 0.052 and IQR of 0.038–0.072. In gastropod families, the median is 0.098, and the IQR is 0.062–0.122. α_b is extremely low in the few brachiopod and barnacle taxa examined with medians of 0.009 and 0.013, and IQRs of 0.006–0.011 and 0.007–0.019, respectively.

Shell mass as a proportion of wet mass (β) is known for fewer microbenthic families and is consistently higher than α , with family-level medians ranging from 0.25 to 0.75 (Fig. 3). Bivalves and gastropods have similar β values with medians of 0.500 and 0.5833, and IQRs of 0.433–0.618 and 0.478–0.705, respectively. Brachiopods have a much lower β on average with a median of 0.358 and IQR of 0.289–0.418, and the one barnacle species tested, the balanid had a median value of 0.781. Interfamily variation in β ranges broadly, 0.4 β , likely due to morphological differences in species, and total water (Fig. 3). For example, Mytilidae displayed a mean β of 0.42; however, when total wet mass is weighed with empty mantel cavities, β increases to 0.63 to match the values reported in other studies like Aldridge & McMahon (1978), highlighting β 's dependence on water.

Accuracy of biomass estimates using live-collected specimens

Using live-collected specimens, the shell-mass model outperformed the linear dimension-based model of estimating organic biomass, especially at large body sizes (Fig. 4). For seven of the nine families tested, the shell-mass model improves the accuracy of biomass estimates, with three advantages. First, the shell-mass model best predicts the measured organic biomass based on the r^2 value (see values reported in each plot within Figure. 4). Second, the shell-mass model remedies a common problem with linear

dimension-based methods, which tend to underestimate the biomass of large individuals owing to shell thickening with age. This underestimation by the length-based model and superior fit by the shell mass-based model across all body sizes is especially apparent in Mytilidae and Veneridae bivalves (Fig. 4). Third, because α & β coefficients are directly tied to anatomical masses, they are more likely to be closely correlated with direct measurement of biomass than linear dimensions, which have to be related to mass via assumptions of body density. As an example of the magnitude of this effect of ontogenetic shell thickening, the Veneridae have low support for the shell-mass model when a single β value is applied. However, if β is increased from 0.59 to 0.65 based on observations that the largest individuals have thicker shells, the r^2 of the shell-mass model increases modestly from 0.392 to 0.410. In many cases, both models would be acceptable estimates, and arguably both should be used together.

The Laqueidae brachiopod and Muricidae gastropods showed lower support for the shell-mass model; the length-based model is preferred. In Laqueidae, the poor fit likely results from this animal's anatomy, with the shells enclosing a large volume of water and very little tissue. As a result, Laqueidae has the lowest α and β of all tested families and is, therefore, most sensitive to the challenge of accurately measuring body water content. Model adherence might be improved by a combination of more samples and greater care in processing: the margin for error in measuring such small quantities is small. In Muricidae, the much larger data set shows strong support for both models, with both accurately determining total wet mass, indicating that larger sample sizes can, in fact, improve shell-mass model fit (Fig. 4).

Biomass estimates using fragmented shell material

The value of γ depends on the ability of an observer to estimate the proportion of shell present consistently and is thus the coefficient presenting the greatest challenge to precision. Overall, shell mass was underestimated by equation 3 when compared to the linear dimension-estimated shell mass (Fig. 5). This underestimate is highlighted by the sum simple residuals of -6.7204 g, and an average simple residual of -0.0234 g. These negative simple residuals reveal a tendency for the observer to underestimate the amount of shell missing and the expected shell thinning during preservation.

In addition to a simple residual, the chi-square error shows that the range of residual error increases with increasing shell height. However,

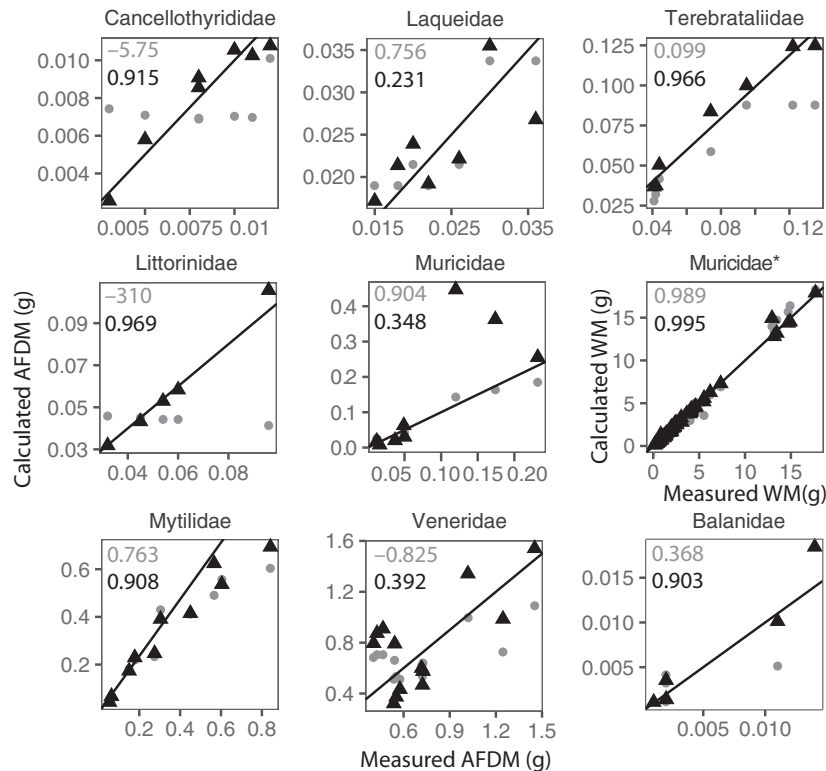


Fig. 4. Biomass (AFDM) of live-collected bivalves calculated using shell mass (Eqn (2), $\gamma = 1$; black triangles) and calculated using a linear dimension (Eqn (4); grey circles) compared with their measured biomass. Each data point denotes a tested specimen. Black lines indicate the ideal, 1:1 relationship of calculated to measured values; numbers in the upper left corner of each graph are r^2 values of the mass- (black) or linear dimension-based (grey) estimates, which are each based on 5–15 specimens and 1–3 species. All plots reflect data produced in this study in units of g AFDM, except for the second Muricidae, which are based on total wet mass (W) data from Palmer (1982). Family-level mean values of α and β derived from data in these graphs are included as ‘Exp.’ data in Fig. 3.

the broadest range of errors is focused around $\gamma = 0.5$, meaning one complete valve was weighed (Fig. 5C). Therefore, this range in chi-square error may reflect natural variability in shell mass with increased size in *Macoma* spp., rather than a predictive error. In this analysis, bivalves had a measurable height, and thus, the variation in thickness from the umbo to commissure is documented and could account for at least some of the variation seen in Figure 5. Surface area estimates could be improved in the future with image processing employing 3-dimensional scans; however, these improvements would likely lead to minimal changes in overall reported biomass. On average, the chi-square error is small (average = 0.065) with natural variation in large individuals creating a large sum chi-square error (18.692), which could not be captured using the linear-dimension model.

Between observers, the estimates of γ were not significantly different, with a Wilcoxon signed-rank test revealing a P -value of 0.383 (Fig. 6). Also, when one observer re-counted the same grab on two separate days, the modes and interquartile ranges were virtually identical with a P -value of 0.522. These results

confirm the reproducibility of this estimate, with reliable precision between observers.

Discussion

The shell-mass model improves upon the traditional linear dimension-based model for estimating biomass (Fig. 4); however, there are three important considerations for the application of this model to future studies.

First, biomass estimates based exclusively on the biomass of shell-producing taxa will only serve as a good proxy of the total biomass of the community to the extent that shelly fauna dominates those communities. Molluscs, and especially bivalves, dominate the biomass of many modern-day benthic communities. However, macrobenthic proportions are highly variable, and molluscs can constitute a small proportion of some macrobenthic communities such that even their larger body sizes cannot compensate for their lack of biomass (e.g. see review in Staff & Powell 1988). Polychaetes are typically numerically dominant as well as most speciose, but in general, the

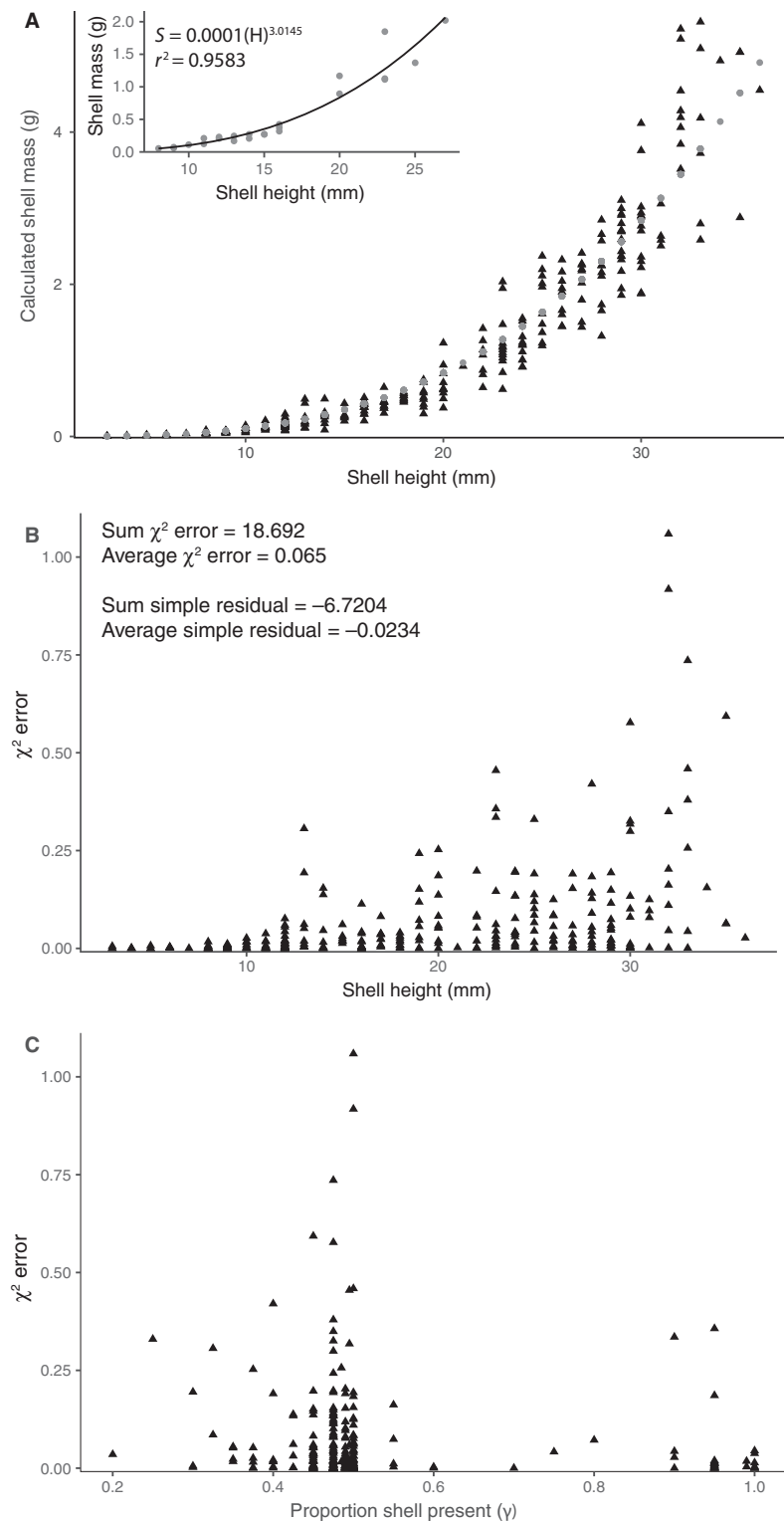


Fig. 5. Reliability of estimates of γ to reconstruct the original, complete shell mass (S) of an individual, using 295 specimens of *Macoma* spp. from death assemblages, each having a measurable dorsal-ventral height. Expected shell mass was calculated using a height-shell mass relationship ($S = 0.0001(H)^{3.0145}$) derived from only complete shells ($\gamma > 95\%$), shown on the inset graph in panel A with the r^2 of this model. (A) The shell mass of an individual was reconstructed twice, once with this height model (grey circles) and once with the shell-mass model (Eqn (3), black triangles). The estimates of original shell mass produced by the two models show strong qualitative overlap. (B) The chi-square error as a function of shell height, showing largest errors at largest shell size. The sum and average of this chi-square error are listed as well as the sum and average of a simple residual (mass estimate – height estimate). (C) chi-square error as a function of γ , to quantify if large chi-square errors are associated with natural shell mass variation or with the γ estimates. The largest errors are found when one complete valve ($\gamma = 0.5$) or shell ($\gamma = 1$) is measured.

molluscs in this 'preservable' community are a significant proportion of the macrobenthic community (Schopf 1978; Staff & Powell 1988; Grebmeier 2012). This variability in the proportional biomass of shelled fauna likely also applied in past seascapes and presents a fundamental challenge to evaluating large-scale spatial and temporal patterns in biomass using only shelled taxa, as long recognized by Staff *et al.* (1985) and Bambach (1993), for example. Improved models for estimating the biomass of *shelled* taxa cannot overcome the fundamental challenge that benthic communities today and, in the past, included many non-shelled metazoans. However, palaeoecological analyses focused exclusively on the numerical abundance or occurrence of only shelled macrobenthos face the same challenge and this bias is not unique to biomass data.

Second, the skeletal mass model depends upon living specimens of target taxa or their congeners to calibrate α and β . This model will thus not be practical with entirely extinct fauna: one must be able to argue on the basis of similar morphology and phylogenetic relationships that modern values can be extrapolated. Importantly, for past representatives of many bivalve families, such an extrapolation is probably warranted, given the narrow ranges of α and β values encountered today (Fig. 3). Moreover, given the fundamental anatomy of articulate brachiopods, with the shell enclosing a bulky feeding structure that is only lightly sheathed in soft tissue, α and β values are likely to remain low as more taxa are tested. The median and IQR values of α and β for modern groups compiled in Fig. 3 thus provide a reasonable initial basis for estimating biomass in fossil assemblages and at the least suggest a way forward, much as meta-analysis and modelling of the reliability of numerical abundance data in modern, mostly molluscan assemblages is proving useful (e.g. review by Kidwell & Tomašových 2013).

Finally, skeletal mass measurements require that the fossil retains its original mineralogy and that shell density is not otherwise affected by diagenesis (e.g. infilling of pores by authigenic precipitates). Unaltered shell material is reasonably assumed for most modern-day death assemblages where shell thinning from dissolution may be more of an issue. Likewise, unaltered shell material is also reasonably assumed for shelly assemblages from many unlithified fossil assemblages (Cenozoic and some Mesozoic records). However, even in these best-case settings, the best procedure will be the estimation of biomass using both the skeletal mass model and a traditional linear dimension-based model to maximize the number of specimens included in the analysis and to benefit from their different taphonomic biases. Linear-

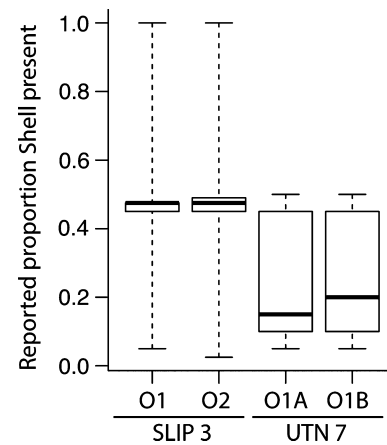


Fig. 6. To test human error in estimating γ using the chart in Figure 2, two operators (O1 and O2) estimated shell completeness of bivalve specimens in the same sample (SLIP3, $N = 127$), and a single operator re-assessed a single sample on two different days (UNT7, $N = 257$). Both samples are diverse and abundant and represent two distinct faunas. Box plots display the median (dark line), interquartile range (box) and the full distribution (whiskers) of γ values reported. The error was insignificant: Wilcoxon signed-rank tests could not reject the null hypothesis for either pair of distributions (O1 vs. O2 $P = 0.383$; O1A vs. O1B $P = 0.522$).

dimension models can include shells that have suffered from mineral replacement, and the shell-outline method developed here can be extended to it.

These caveats notwithstanding, the skeletal mass model developed here represents a significant improvement to estimating biomass. The harshest critique of the reliability of fossil biomass models has been their inability to include fragmented and colonial individuals (Waselkov 1987; Forcino *et al.* 2010). Although not explicitly tested here, this model also shows promise in overcoming the inability of biovolume models to measure colonial taxa biomass and the difficulty of comparing abundances of solitary and colonial organisms, also known as the colonial conundrum (Forcino & Leighton 2010). Estimating γ for a colonial organism is not strictly speaking possible. However, by applying Eqn 2 with $\gamma=1$, an investigator could find a minimum biomass estimate. For solitary individuals, the human error associated with γ is small (Figs 5 and 6). Adding γ to the model expands what is 'usable' in a fossil assemblage by including fragmented solitary individuals, increasing the reliability of the biomass estimate. The results here thus suggest that an investigator can take advantage of much of the preserved shelly community.

Conclusions

Most critiques of traditional biomass estimates with linear-dimension models can be mitigated or

overcome with the skeletal mass model demonstrated here. The capability to find biomass in the fossil record is no longer a question of reliability and is now a question of which currency is best suited to achieve the research goal. Because carbon and nutrients are exchanged at every scale of biological organization, a reliable means of estimating fossil biomass can help bridge the gap between modern-day and ancient ecosystems with the reconciliation of the current community assemblages and ancient averaged data. Biomass of a fossil represents the net secondary productivity of that animal from birth until death, and fossil assemblages include individuals that lived and died throughout the accumulation window, making biomass fundamentally time-averaged and complementary but different from modern estimates (Staff *et al.* 1985, 1986; Powell *et al.* 2001). Biomass as a metric in palaeoecology is currently being used to re-examine fundamental questions such as constraints on body size evolution (e.g. Lawton 1990; Payne & Finnegan 2006; Heim *et al.* 2015), and the trophic structure of recent and ancient assemblages (Walker 1972; Powell *et al.* 2001). In addition, biomass can be used to evaluate the temporal consistency of modern observed patterns through time, such as the gradient of increasing benthic biomass (gC/m^2 ; Wei *et al.* 2010; Powell & Mann 2016); the established relationships between benthic productivity and nutrient availability (Grebmeier *et al.* 1988; Martin 1996); and the patterns and processes of ecological succession (Nilsson & Rosenberg 2000). Each question and pattern listed here could not be answered without the added insights of biomass, production, and energetics and suggest new paradigms could arise from the fossil record and palaeoecological approaches.

Acknowledgements. – This research was supported by the Department of the Geophysical Sciences and the Gurley fund for Paleobiologic Research at the University of Chicago. I would like to thank Michael LaBarbera, Michael Foote and Gerald Olack (University of Chicago) for guidance in the laboratory and feedback on analytical methods; Jacqueline Grebmeier at the Chesapeake Biological Laboratory of the University of Maryland Center for Environmental Science for providing samples of death assemblages from the North Pacific Arctic, and for discussion of biomass in the living communities there; Nicole Bitler and Michael LaBarbera for donations of live-collected molluscs and brachiopods; Kenzo Esquivel and Eva Haraldsdottir for assistance in the laboratory; and Susan Kidwell, David Jablonski, Albert Coleman, Cathy Pfister, Lee Cooper and Adam Tomašových for feedback on the manuscript. I also thank two anonymous reviewers for constructive and valuable feedback on the manuscript. Data for Figs 3 and 4 are available at url: <https://figshare.com/s/255e587a61fa9a2bbb32>.

References

- Aldridge, D.W. & McMahon, R.F. 1978: Growth, fecundity, and bioenergetics in a natural population of the Asiatic freshwater clam, *Corbicula manilensis philippi*, from North Central Texas. *Journal of Molluscan Studies* 44, 49–70.
- Allmon, W.D. & Martin, R.E. 2014: Seafood through time revisited: the Phanerozoic increase in marine trophic resources and its macroevolutionary consequences. *Paleobiology* 40, 256–287.
- Bambach, R.K. 1993: Seafood through time: changes in biomass, energetics, and productivity in the marine ecosystem. *Paleobiology* 19, 372–397.
- Brey, T. 2001: *Population Dynamics in Benthic Invertebrates. A Virtual Handbook, Version 01.2*. Alfred Wegener Institute for Polar and Marine Research, Germany, <http://www.thomas-brey.de/science/virtualhandbook>.
- Brey, T., Rumohr, H. & Ankar, S. 1988: Energy content of macrobenthic invertebrates: general conversion factors from weight to energy. *Journal of Experimental Marine Biology and Ecology* 117, 271–278.
- Brey, T., Müller-Wiegmann, C., Zittler, Z.M. & Hagen, W. 2010: Body composition in aquatic organisms—a global data bank of relationships between mass, elemental composition and energy content. *Journal of Sea Research* 64, 334–340.
- Brown, J.H., Gillooly, J.F., Allen, A.P., Savage, V.M. & West, G.B. 2004: Toward a metabolic theory of ecology. *Ecology* 85, 1771–1789.
- Cameron, C., Cameron, I. & Paterson, C. 1979: Contribution of organic shell matter to biomass estimates of unionid bivalves. *Canadian Journal of Zoology* 57, 1666–1669.
- Cravo, A., Bebianno, M. & Foster, P. 2004: Partitioning of trace metals between soft tissues and shells of *Patella aspera*. *Environment International* 30, 87–98.
- Davoult, D., Harlay, J. & Gentil, F. 2009: Contribution of a dense population of the brittle star *Acrocnida brachiata* (Montagu) to the biogeochemical fluxes of CO_2 in a temperate coastal ecosystem. *Estuaries and Coasts* 32, 1103–1110.
- Finnegan, S. 2013: Quantifying seafood through time: counting calories in the fossil record. *The Paleontological Society Papers* 19, 21–50.
- Forcino, F.L. & Leighton, L.R. 2010: Determining the most robust method of measuring disaggregated fossil material: the colonial conundrum. In: 2010 GSA Denver Annual Meeting, Geological Society of America Abstracts with Programs 42, 257, Geological Society of America, Denver.
- Forcino, F.L., Stafford, E.S., Warner, J.J., Webb, A.E., Leighton, L.R., Schneider, C.L., Michlin, T.S., Palazzolo, L.M., Morrow, J.R. & Schellenberg, S.A. 2010: Effects of data categorization on paleocommunity analysis: A case study from the Pennsylvanian Finis Shale of Texas. *Palaos* 25, 144–157.
- Forcino, F.L., Leighton, L.R., Twerdy, P. & Cahill, J.F. 2015: Re-examining sample size requirements for multivariate, abundance-based community research: when resources are limited, the research does not have to be. *PLoS ONE* 10, e0128379.
- Glover, C.P. & Kidwell, S.M. 1993: Influence of organic matrix on the post-mortem destruction of molluscan shells. *The Journal of Geology* 101, 729–747.
- Grebmeier, J.M. 2012: Shifting patterns of life in the Pacific Arctic and sub-Arctic seas. *Annual Reviews in Marine Science* 4, 63–78.
- Grebmeier, J.M., McRoy, C.P. & Feder, H.M. 1988: Pelagic-benthic coupling on the shelf of the northern Bering and Chukchi Seas. I. Food supply source and benthic biomass. *Marine Ecology Progress Series* 48, 57–67.
- Guedes, L.M.L., Fiori, A. & Diefenbach, C.D.C. 1981: Biomass estimation from weight and linear parameters in the apple snail, *Ampullaria canaliculata* (Gastropoda: Prosobranchia). *Comparative Biochemistry and Physiology Part A: Physiology* 68, 285–288.
- Heim, N.A., Knope, M.L., Schaal, E.K., Wang, S.C. & Payne, J.L. 2015: Cope's rule in the evolution of marine animals. *Science* 347, 867–870.
- Kidwell, S.M. & Tomašových, A. 2013: Implications of time-averaged death assemblages for ecology and conservation biology. *Annual Review of Ecology, Evolution, and Systematics* 44, 539–563.

- Lawton, J. 1990: Species richness and population dynamics of animal assemblages. Patterns in body size: abundance space. *Philosophical Transactions: Biological Sciences* 330, 283–291.
- Lejart, M., Clavier, J., Chauvaud, L. & Hily, C. 2012: Respiration and calcification of *Crassostrea gigas*: contribution of an intertidal invasive species to coastal ecosystem CO₂ fluxes. *Estuaries and Coasts* 35, 622–632.
- Lyman, R.L. 2008: *Quantitative Paleozoology*. Cambridge University Press, Cambridge.
- Martin, R.E. 1996: Secular increase in nutrient levels through the Phanerozoic: implications for productivity, biomass, and diversity of the marine biosphere. *Palaos* 11, 209–219.
- McDonald, J.H. 2009: *Handbook of Biological Statistics*. 2nd edn. Sparky House Publishing, Baltimore.
- Nilsson, H.C. & Rosenberg, R. 2000: Succession in marine benthic habitats and fauna in response to oxygen deficiency: analysed by sediment profile-imaging and by grab samples. *Marine Ecology Progress Series* 197, 139–149.
- Palmer, A.R. 1982: Growth in marine gastropods. A non-destructive technique for independently measuring shell and body weight. *Malacologia* 23, 63–74.
- Palmerini, P. & Bianchi, C. 1994: Biomass measurements and weight-to-weight conversion factors: a comparison of methods applied to the mussel *Mytilus galloprovincialis*. *Marine Biology* 120, 273–277.
- Payne, J. & Finnegan, S. 2006: Controls on marine animal biomass through geological time. *Geobiology* 4, 1–10.
- Powell, E.N. & Mann, R. 2016: How well do we know the infaunal biomass of the continental shelf? *Continental Shelf Research* 115, 27–32.
- Powell, E. & Stanton, J. 1985: Estimating biomass and energy flow of molluscs in palaeo-communities. *Paleontology* 28, 1–34.
- Powell, E.N., Staff, G.M., Stanton, R.J. & Callender, W.R. 2001: Application of trophic transfer efficiency and age structure in the trophic analysis of fossil assemblages. *Lethaia* 34, 97–118.
- Ricciardi, A. & Bourget, E. 1998: Weight-to-weight conversion factors for marine benthic macroinvertebrates. *Marine Ecology Progress Series*, 171, 245–251.
- Rumohr, H., Brey, T. & Ankar, S. 1987: *A Compilation of Biometric Conversion Factors for Benthic Invertebrates of the Baltic Sea* 9. Institut für Meereskunde, Kiel.
- Schopf, T.J. 1978: Fossilization potential of an intertidal fauna: Friday Harbor, Washington. *Paleobiology* 4, 261–270.
- Staff, G.M. & Powell, E. 1988: The paleoecological significance of diversity: the effect of time averaging and differential preservation on macroinvertebrate species richness in death assemblages. *Palaeogeography, Palaeoclimatology, Palaeoecology* 63, 73–89.
- Staff, G., Powell, E.N., Stanton, R.J. & Cummins, H. 1985: Biomass: is it a useful tool in paleocommunity reconstruction? *Lethaia* 18, 209–232.
- Staff, G.M., Stanton, R. Jr, Powell, E. & Cummins, H. 1986: Time-averaging, taphonomy, and their impact on paleocommunity reconstruction: death assemblages in Texas bays. *GSA Bulletin* 97, 428–443.
- Steimle, F. & Terranova, R.J. 1985: Energy equivalents of marine organisms from the continental shelf of the temperate North-west Atlantic. *Journal of Northwest Atlantic Fishery Science* 6, 117–124.
- Stoker, S.W. 1978: *Benthic Invertebrate Macrofauna of the Eastern Continental Shelf of the Bering and Chukchi Seas*, 259 pp. Unpublished PhD Thesis, University of Alaska, Fairbanks.
- Thorson, G. 1957: Bottom communities (sublittoral or shallow shelf). *Geological Society of America Memoirs* 67, 461–534.
- Waldbusser, G.G., Powell, E.N. & Mann, R. 2013: Ecosystem effects of shell aggregations and cycling in coastal waters: an example of Chesapeake Bay oyster reefs. *Ecology* 94, 895–903.
- Walker, K.R. 1972: Trophic analysis: a method for studying the function of ancient communities. *Journal of Paleontology* 46, 82–93.
- Waselkov, G.A. 1987: Shellfish gathering and shell midden archaeology. *Advances in Archaeological Method and Theory* 10, 93–210.
- Wei, C.L., Rowe, G.T., Escobar-Briones, E. et al. 2010: Global patterns and predictions of seafloor biomass using random forests. *PLoS ONE* 5, e15323.