A generalised volumetric method to estimate the biomass of photographically surveyed benthic megafauna

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ABSTRACT

Biomass is a key variable for understanding the stocks and flows of carbon and energy in the environment. The quantification of megabenthos biomass (body size ≥ 1 cm) has been limited by their relatively low abundance and the difficulties associated with quantitative sampling. Developments in robotic technology, particularly autonomous underwater vehicles, offer an enhanced opportunity for the quantitative photographic assessment of the megabenthos. Photographic estimation of biomass has typically been undertaken using taxon-specific length-weight relationships (LWRs) derived from physical specimens. This is problematic where little or no physical sampling has occurred and/or where key taxa are not easily sampled. We present a generalised volumetric method (GVM) for the estimation of biovolume as a predictor of biomass. We validated the method using fresh trawl-caught specimens from the Porcupine Abyssal Plain Sustained Observatory (northeast Atlantic), and we demonstrated that the GVM has a higher predictive capability and a lower standard error of estimation than the LWR method. GVM and LWR approaches were tested in parallel on a photographic survey in the Celtic Sea. Among the 75% of taxa for which LWR estimation was possible, highly comparable biomass values and distribution patterns were determined by both methods. The biovolume of the remaining 25% of taxa increased the total estimated standing stock by a factor of 1.6. Additionally, we tested inter-operator variability in the application of the GVM, and we detected no statistically significant bias. We recommend the use of the GVM where LWRs are not available, and more generally given its improved predictive capability and its independence from the taxonomic, temporal, and spatial, dependencies known to impact LWRs.

1. Introduction

A census of biomass on Earth is important to understanding both the structure and the functioning of the biosphere (Bar-On et al., 2018). Population and assemblage biomass, together with individual body size (mass), are generally seen to be critical variables in the assessment of the stocks and flows of mass and energy in marine ecosystems (e.g. Tomlinson et al., 2014). These stocks and flows influence the primary ecosystem goods and services that the marine environment provides, and in turn, monitoring their status is likely to be essential to achieving the corresponding sustainable development goals (United Nations General Assembly, 2015). In the following, we consider the need for benthic biomass data, the current synthesis of global megabenthos data (large fauna living on or near the seafloor), and the suitability of the currently predominant biomass estimation method.

1.1. Biomass as an essential variable

The Framework for Ocean Observing (e.g. Lindstrom et al., 2012) indicates a region of overlap between the essential biodiversity variables (EBVs) of the global biodiversity observing system (GEO BON; Kissling et al., 2018) and the essential ocean variables (EOVs) of the global ocean observing system (GOOS; Muller-Karger et al., 2018), within which the variable body mass lies. Body mass directly, or indirectly, features in several EBVs: (a) population abundance (as a proxy for numerical abundance); (b) population structure by age/size class; (c) body mass; (d) physiological traits (as a key predictor of metabolism and related traits; e.g. Peters, 1983); and (e) secondary productivity (as a key predictor; e.g. Banse, 1980). Assessment of these EBVs is considered relevant to Aichi Biodiversity Targets 4–12 and 14–15 for the maintenance and the restoration of biological ecosystems by 2020 (GEO BON, 2011; Pereira et al., 2013).

Recent examinations of EOVs (e.g. Constable et al., 2016; Miloslavich et al., 2018; Levin et al., 2019) have made frequent references to abundance and to biomass variables, and occasional references to body-mass spectra, in connection with zooplankton, fish, and benthic invertebrates. The GOOS currently recognises stock assessments of marine biota groups, including benthic invertebrate stocks, as 'emerging' EOVs (www.goosocean.org, accessed September 2019). The deep ocean observing strategy (DOOS; Levin et al., 2019) recommends the consideration of EOVs and of 'emerging' EOVs, with the addition of individual body size and of sponge-habitat cover (www.deepoceanobserving.org, accessed September 2019). There is general agreement in the identification of biomass as a key variable in the EBVs by GEO BON, and in the biology and ecosystem EOVs by GOOS and DOOS.

Given the central roles that individual body size and total biomass play in the stocks and flows of mass and energy through marine ecosystems, it seems clear that they should be established as EOVs. Selection as an essential variable is not determined by the variable's perceived 'value' or 'need' alone. The expert panels and other researchers that consider candidate EOVs necessarily give regard to many factors (e.g. Miloslavich et al., 2018); key among these are likely to be: (a) impact, i.e. scientific and societal relevance; (b) feasibility, i.e. monitoring scalability and practicality; and (c) cost effectiveness, i.e. scientific and operational capacity. Given that body mass already features in multiple EBVs of the GEO BON, its impact for ecological research seems clear. It is therefore timely and of particular significance, for both the scientific community and conservation practitioners, to establish a method for the estimation of individual body size and total biomass that (i) has general, broad-scale, application, (ii) can readily be adopted for use in multiple environments by a wide range of users, and (iii) can be achieved using readily available existing technologies.

1.2. Existing benthic megafauna biomass data

In the marine environment, recent field studies (Kelly-Gerreyn et al., 2014; Labra et al., 2015; Laguionie-Marchais et al., in review) and theoretical considerations (Bett, 2013) have suggested that total estimated seafloor biomass increases with the mean, or maximum, body size of the organisms included in the study. In effect, the largest organisms present, e.g. benthic megafauna (megabenthos; ≥1 cm body size), contribute substantially to the 'true' total standing stock biomass in the system (Bett, 2019). Seafloor megafaunal assemblages are often assessed by trawl sampling (e.g. Gage and Bett, 2005); however, this approach is typically limited to sedimentary habitats and is likely to be semi-quantitative at best (e.g. McIntyre, 1956; Uzmann, 1977; Bett et al., 2001). Moreover, trawling, be it for scientific sampling or commercial fishing purposes, is a rather destructive process (e.g. Huvenne et al., 2016) and thus should be avoided if possible; a factor that may be of concern in time-series studies (e.g. Billett et al., 2010). In response to these limitations, there has been a general increase in the use of seafloor photography to quantify megabenthos assemblages (Durden et al., 2016c), which has recently accelerated with the development of autonomous underwater vehicles (AUVs; Wynn et al., 2014; Jones et al., 2019) and their use in quantitative seafloor ecology (Morris et al., 2016; Simon-Lledó et al., 2019). AUV-based seafloor photography enables the rapid quantitative survey of large areas that can comprise multiple habitats in a consistent and non-destructive manner (e.g. Morris et al., 2014; Marzinelli et al., 2015; Milligan et al., 2016; Benoist et al., 2019).

Wei et al. (2010) provide a major compilation and assessment of global benthic biomass (from bacteria to megabenthos) that is widely cited (129 Web of Science Core Collection citations, May 2019) and that has been used as the basis of other major works, e.g. to predict future trends of seafloor biomass in response to climate change (Jones et al., 2014; 58 Web of Science Core Collection citations, May 2019). However, the megabenthos biomass data synthesised by Wei et al. (2010)

encompasses records based on bottom-trawl catches and on photographic surveys, potentially introducing mismatches in the spatial scale observed and in the body sizes and the taxonomic groups assessed. We examine the possible significance of this methodological variation in our evaluation of available methods below.

1.3. Photographic estimation of individual biomass

Generating quantitative ecological data from large sets of seafloor photographs poses a number of challenges including specimen detection and identification, biomass estimation, and data standardization. Durden et al. (2016a) provide a brief review of existing methodology that essentially comprise two options: (i) calculating the product of numerical density and of a representative value for the individual biomass of a particular taxon, or (ii) the use of a taxon-specific length-weight relationship (LWR) approach. Both methods require access to existing morphometric data on the taxa of interest, which are frequently unavailable in poorly studied geographic regions, in particular deep-water environments, or for the attached fauna of hard-substratum habitats.

Nevertheless, the LWR approach is commonly employed in both the analyses of photographic surveys (e.g. Durden et al., 2015) and of trawl catches (e.g. Robinson et al., 2010). These conversions are typically expressed as an allometric equation of the form:

$$M_E = a \times SL^b \qquad (1)$$

where M_E is estimated body mass, SL is a defined standard linear body dimension, and a and b are taxon-specific constants obtained by log-log regression of measured body mass on SL, and consequently require adequate prior data for the taxon in question (e.g. Durden et al., 2016a). These two constants are an expression of life history and local environmental settings (i.e. ecological factors affecting individual metabolism; e.g. Peters, 1983). In biological terms, the b parameter (or allometry coefficient) is indicative of the rate of weight gain relative to growth in length (see Eq. (2) below). The LWR method is attractive for both its simplicity and its ability to generate biomass estimates for individual specimens, such as are required in the study of individual-based body-size spectra (Edwards et al., 2017; Laguionie-Marchais et al., in review) or any research involving the structuring role of body size in ecosystems (e.g. Sewall et al., 2013; Lewis et al., 2018; Durden et al., 2019).

Durden et al. (2016a) accessed a database of some 47,000 specimens of megabenthos collected by otter trawl from the Porcupine Abyssal Plain Sustained Observatory (PAP-SO; 4850 m water depth, northeast Atlantic) to produce LWRs for 34 morphotypes (species/species groups/higher taxa; e.g. Althaus et al., 2015). A typical otter-trawl catch from the PAP-SO returns specimens from between 60 and 80 morphotypes, and the current morphotype catalogue for photographic studies in the area has some 70 morphotypes (Durden et al., this issue; Hosking et al., this issue). However, despite the high research effort at PAP-SO (Billett and Rice, 2001; Lampitt et al., 2010; Hartman et al., 2012; Guest editors, this issue), LWRs are only available for approximately half of the taxa present. Similarly, Robinson et al. (2010) undertook a major beam-trawl survey of 283 stations in the North Sea, encountering 497 benthic fish and invertebrate taxa, from which they were able to produce LWRs for 216 taxa, i.e. approximately half of the taxa present. Of those 216 LWRs, only 95 were based on 50 or more specimens. Nine of these invertebrate species were sufficiently numerous and widespread in their geographic distribution to analyse temporal (year of sampling) and spatial (north or south of the 50 m isobath) variation in the LWRs. The authors detected statistically significant temporal and/or spatial variations in the LWRs of seven of those species (see also Stoffels et al., 2003).

Potential temporal (and spatial) variation in the a and the b parameters of LWRs are linked to the concept of condition factor (or condition index), as frequently implemented in studies of fish populations

(e.g. Froese, 2006). The condition factor is essentially a ratio of measured specimen body mass to the mass predicted from the specimen's length. Froese (2006) provides a formula for calculating relative weight (W_{rm}) as:

$$W_{rm} = 100 \times W/a_m L^{b_m} \qquad (2)$$

where W is specimen weight, L is specimen length, and a_m and b_m are the geometric means of the available LWR parameters. In terms of statistical significance, the number of specimens used to calculate a LWR can be expected to impact its reliability. We examine the possible significance of systematic variation in condition factor in our evaluation of available methods below.

1.4. The need for a generalised method

LWRs are potentially subject to systematic, temporal and spatial, variation, and may be highly taxon-specific. Consequently, the use of LWRs out of temporal, spatial, or taxonomic, context may result in substantial systematic error. More fundamentally, taxon-specific LWRs simply do not exist for the vast majority of megafaunal species, imposing an immediate severe limitation on the general application of this approach. Environmental assessments, particularly in relation to deep-sea mineral resource exploitation, are now regularly being conducted in very poorly known areas where physical sampling of the megafauna is rare or absent (e.g. Gates et al., 2017; Durden et al., 2018; Stratmann et al., 2018), demonstrating a growing need for a more tractable method of taxon-independent biomass estimation.

Consequently, we have developed a taxon-independent method for the estimation of biovolume, from geometric considerations of photographed specimens, as a proxy for biomass. Similar biovolume-based approaches are well established for small organisms (microbes: Saccà, 2017; phytoplankton: Jiménez et al., 1987; Hillebrand et al., 1999; Sun and Liu, 2003; zooplankton: Alcaraz et al., 2003; Mustard and Anderson, 2005; copepods, nematodes: Baguley et al., 2004; Di Mauro et al., 2011; Jung et al., 2012; Moore et al., 2013; Mazurkiewicz et al., 2016; gastropods: McClain, 2004) and fossil invertebrates (Novack-Gottshall, 2008). Briefly, these approaches select a geometric form to represent approximately the biovolume of a given taxon, then make the measurements necessary to estimate the volume of the selected geometric form. Further conversion of biovolume to units of (fresh) wet weight mass, carbon mass (C), or energy (e.g. joule) can be achieved via established factors (e.g. Brey et al., 2010).

In this contribution, we describe and test a generalised volumetric method (GVM) for the estimation of megafaunal specimen biovolume, as an estimator of biomass, from photographic observations. We first validated the method against measured specimen mass and volume using a collection of fresh trawl-caught specimens from the PAP-SO site. The full methodology was then trialled by two operators in a case study of benthic ecology based on a large photographic dataset derived from AUV surveys on the Celtic Shelf (100 m water depth, northeast Atlantic), where both sedimentary and hard substratum habitat types occur. Comparative assessments of the conventional taxon-specific LWR approach and the proposed taxon-independent GVM are provided, together with an assessment of inter-operator variation in biovolume and biomass estimation.

2. Materials and methods

2.1. Evaluation of current methods

2.1.1. Field methods

To evaluate the influence of field method, megabenthos biomass data (invertebrates and fish) from the Wei et al. (2010) dataset, limited to deep-sea records (water depth > 200 m), were separated into trawl catches and photographic surveys. A general linear model (LM) of log (biomass) on water depth by method of biomass estimation was developed using the Minitab software package (v18.1; Minitab, Inc.).

2.1.2. Length-weight relationship (LWR) method

To examine the characteristics of the LWR method for predicting individual biomass, we examined morphometric data for the large holothurian *Psychropotes longicauda* ($n = 984$) from 15 trawl catches spanning seven research cruises (different years) to the PAP-SO. This species is a biomass dominant at PAP (e.g. Billett et al., 2010) and easily identified in both trawl catches and seafloor photographs. Corresponding LWRs were examined by linear regression (preserved wet weight ~ standard body length), and temporal variation (between individual catches and years) by LM (preserved wet weight ~ standard body length × trawl or × year), as performed in Minitab. The relative weight of *P. longicauda* specimens was calculated using Eq. (2) with the a_m and b_m parameters taken to be the cruise (year) values. Non-parametric tests (Spearman's rank correlation and Mood's median test; Minitab) were used to further examine variations in relative weight.

2.2. A generalised volumetric method (GVM)

The GVM models specimen body volume as a cylinder, and therefore it requires two defining measurements that correspond to the radius (measured as the diameter) and to the length of an equivalent cylindrical object. This approach represents a much simplified approximation of the full range of body forms exhibited by benthic megafauna. Consequently, the method requires the user to compress conceptually the specimen into a cylinder of approximately equivalent volume. **(1)** The user must first choose the most appropriate axis of rotation for the cylinder; this will become the dimension along which length is measured. The choice of an 'appropriate' axis is essentially determined by the general body plan and the orientation of the photographed specimen (Fig. 1). **(2)** The next choice is an appropriate equivalent cylindrical diameter (*ECD*) perpendicular to the axis of rotation, i.e. given the chosen axis of rotation, what is the most appropriate representative diameter for a conceptually compressed cylinder in that orientation? **(3)** Finally, given that choice of *ECD*, what is the most appropriate equivalent cylindrical length (*ECL*) that will best estimate the volume of the cylinder (Figs. 1 and 2)? The estimated specimen biovolume (V_E) is then calculated as:

$$V_E = \pi \times (ECD/2)^2 \times ECL \qquad (3)$$

The process is simplest to conceive in the case of vermiform organisms (Fig. 2a and b); however, it is readily translated to a wide range of morphologies (Fig. 2c–l). The method is necessarily subjective in that the measurements are not made between distinct morphological features, but they are instead aimed at the most effective volumetric
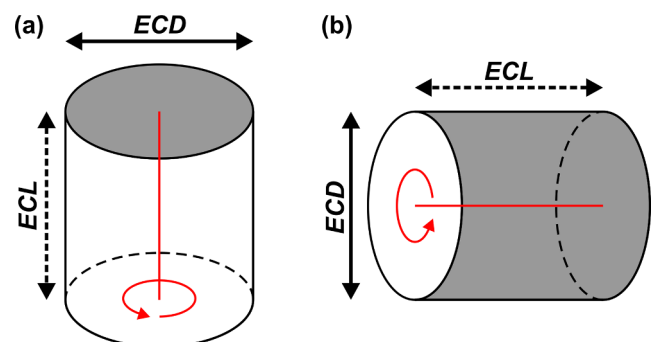

Fig. 1. Body dimensions of a conceptualised cylindrical specimen as observed from (a) a vertical or (b) a horizontal orientation. The observed body surface (shaded), the axis of orientation (red line), the equivalent cylindrical diameter (*ECD*; solid dimension line), and the equivalent cylindrical length (*ECL*; dashed dimension line) are illustrated. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)
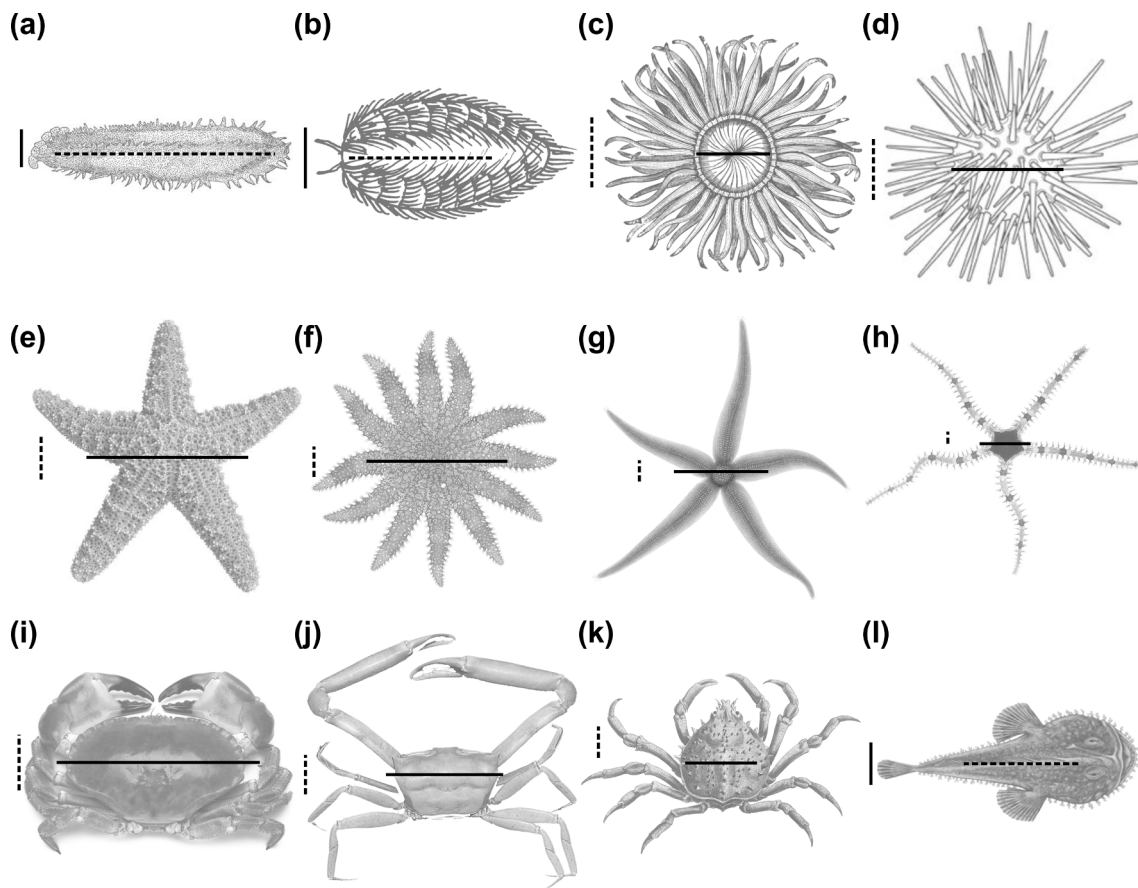
Fig. 2. Examples of the application of generalised volumetric method measurements to a range of benthic megafauna body forms. The corresponding equivalent cylindrical diameter (*ECD*; solid line) and equivalent cylindrical length (*ECL*; dashed line) measurements are illustrated. (a) Holothuroidea. (b) Polychaeta. (c) Anthozoa. (d) Echinoidea. (e–g) Asteroidea. (h) Ophiuroidea. (i–k) Brachyura. (l) Actinopterygii.

representation. The method is, therefore, (*recommendation 1*) best implemented by users with zoological knowledge of the taxa involved and, preferably, (*recommendation 2*) with experience in directly handling comparable physical specimens. It is simplistic but affords considerable flexibility in practical application, enabling the user to deal with varying specimen orientation and/or partially obscured specimens. From experience, (*recommendation 3*) our primary advice to potential users is to retain a simple focus on the objective of estimating the tissue biovolume of the specimen in question, including its appendages if they make an appreciable contribution to the organism's volume. The user should avoid the temptation of making a 'standard measurement' if that has been their previous practice, and treat each specimen encountered as a new case. The method draws on the user's prior knowledge and experience of three-dimensional morphology; consequently, that knowledge is a prerequisite for successful operation.

The GVM is readily adapted to colonial, encrusting, or morphologically plastic, forms (e.g. Ascidiacea, Bryozoa, Cnidaria, Porifera). In colonial forms, the user can opt to estimate the colony as a unit, or to make estimates for the unitary components (zooids, polyps). For example, with close-encrusting colonies and Porifera, the user can estimate an *ECD* to best represent the areal extent of the subject and then estimate an *ECL* to best represent the typical thickness of the corresponding layer of biological tissue. With erect colonial Cnidaria (e.g. Octocorallia), the user can (a) estimate the biovolume of each single polyp with a representative contribution of connecting tissue; (b) estimate the biovolume of a single polyp with a representative contribution of connecting tissue, and apply a multiplier for the number *n* of polyps in the colony (i.e. *ECL* is replaced by *ECL* × *n* in Eq. (3)); or (c) estimate an *ECD* representative of stem tissue thickness and then estimate an *ECL*

that represents the total length of the tissue-bearing stem. A very similar approach can be applied to branching Porifera. With other sponge growth forms, the user can readily adopt similar methods, for example: (i) laminar, *ECD* to represent the plate area, *ECL* to represent the plate tissue thickness; (ii) cup/goblet/barrel forms, *ECD* to represent one-half of the outer surface of the cup, *ECL* to represent double the cup tissue thickness. Again, our primary advice to potential users is to retain a simple focus on the objective of estimating the tissue biovolume of the specimen in question (*recommendation 3*).

2.3. Method validation with physical specimens

Fresh specimens of benthic invertebrate megafauna and demersal fish were collected from the Porcupine Abyssal Plain Sustained Observatory site (PAP-SO; 48°50′N 016°30′W) at 4850 m water depth (Hartman et al., 2012), using a semi-balloon otter trawl during the RRS *Discovery* cruises DY050 in 2016 (Stinchcombe, 2017) and DY077 in 2017 (Lampitt, 2017). In total, 206 intact specimens were selected for direct physical measurement on board and subsequent indirect photographic body-size measurement. The test specimens were chosen to represent a wide range of body shapes, sizes (five orders of magnitude), taxonomic identities (six phyla, 34 taxa; Appendix A in Supplementary Material), and ecological characteristics (deposit feeder, filter feeder, predator, scavenger, mobile, sessile).

2.3.1. Direct measurement of specimens

Blotted individual fresh wet weight (fwwt) biomass (M_M) was recorded to the nearest 0.1 g using a motion-compensated electronic balance (POLS S-182 Marine Onboard Scale, Lorrimar Weighing Ltd.).

Fresh biovolume (V_M) was measured by displacement using a measuring cylinder suited to the specimen size (100, 250, 1000, or 2000 mL) and recorded to the nearest 0.5, 1.0, 5.0, or 10.0 mL, respectively. Specimen biomass and biovolume ranged c. 0.5–1225 g and 0.5–1210 mL, respectively.

2.3.2. Indirect measurement of specimens

Each fresh specimen was then photographed (Fujifilm FinePix F550EXR) from above (i.e. high oblique, near perpendicular view), in a position to match the typical view obtained from seafloor AUV-survey photographs (e.g. dorsal view of squat lobster with tail folded beneath body, dorsal view of shrimp, oral view of anemone; Fig. A.1). Photograph-derived body-size measurements were then made at 0.5 mm resolution, typical of seafloor survey photographs (e.g. Morris et al., 2014), via image analysis software (Image-Pro Plus, v7.0, Media Cybernetics Inc.). Three body dimensions were recorded from each specimen: (i) GVM equivalent cylindrical diameter (*ECD*), (ii) GVM equivalent cylindrical length (*ECL*), and (iii) LWR standard linear body dimension (*SL*), as employed in previously established PAP-SO taxon-specific LWRs (i.e. Durden et al., 2016a). The *ECD* and *ECL* measurements were converted to estimated biovolume (V_E) using Eq. (2), i.e. the proposed GVM approach. The *SL* measurement was converted to fresh wet weight biomass (M_E) using Eq. (1), i.e. the LWR method detailed by Durden et al. (2016a).

2.3.3. Analytical approach

Relationships between measured and estimated specimen mass and volume were examined by regression. We primarily based our assessment on the predictive results of model I ordinary least squares (OLS) regression, as implemented in Minitab (Sokal and Rohlf, 1995), on the premise that our focus was the prediction of mass from estimated volume (V_E), or from standard length (*SL*), and that the test specimens were deliberately selected (i.e. intact) rather than randomly sampled from the trawl catches. We carried out OLS regressions on the native variables and on their transformation to natural logarithms to acknowledge potential inhomogeneity of variance. In reporting regression results, we have included the 'Predicted R^2' statistic (Minitab, 2013); this is based on a leave-one-out cross-validation approach and assesses how well the model predicts new observations (see e.g. Allen, 1971). In addition, we also carried out model II regressions (Legendre and Legendre, 1998) that are suited to the assessment of functional relationships where both variables are measured with error, and where the focus is on the symmetric relationship between the two variables, rather than the asymmetric case of predicting one from the other. We implemented two forms of model II regression: (i) ranged major axis (RMA) using the 'lmodel2' package (v1.7–3; Legendre, 2018), and (ii) standardised major axis (SMA) using the 'smatr' package (v3; Warton et al., 2012), in the R environment (v3.3.2; R Core Team, 2016). These various regression techniques are extensively discussed by Warton et al. (2006) and Legendre (2018).

2.4. Method trial in a photographic case study

Seafloor images were obtained from three shelf-sea locations in the Celtic Sea, northeast Atlantic, c. 100 m water depth (Thompson et al., 2017), using the AUV *Autosub3* (McPhail et al., 2009) during the RRS *Discovery* cruise DY034 in 2015 (Ruhl, 2016). The AUV was programmed to survey at a target altitude of 2.5 m above the seafloor, yielding a nominal resolution of c. 0.5 mm per pixel. The optical axis of the camera was approximately perpendicular to the seafloor, with the AUV's pitch and roll angles recorded for subsequent image processing. General field method and subsequent image processing and assessment were as described by Morris et al. (2014, 2016), with data generated from 4160 images, representing c. 4000 m² of seafloor.

2.4.1. Image analysis

All benthic invertebrate megafauna and demersal fish (≥ 1 cm body size) observed were counted and identified to the lowest taxonomic or morphotype unit (Table A.1). As defined above, three body dimensions were recorded per specimen: *ECD*, *ECL*, and *SL*. Where specimens could not be assigned to a taxonomic unit or morphotype, they were recorded as indeterminate ($< 1\%$ of specimens) and excluded from subsequent analyses. Specimen body-size measurements were converted to estimated volume (V_E) using Eq. (2), i.e. the GVM approach, and to estimated mass (M_E) using Eq. (1), i.e. the LWR approach, where possible (via conversion factors obtained from the literature; Coull et al., 1989; Richardson et al., 2000; Robinson et al., 2010; Silva et al., 2013; Durden et al., 2016a). The biovolume of all identifiable specimens recorded was estimated using the GVM; however, LWRs were only available for c. 75% of the taxa encountered. To enable direct comparison of the two methodologies, we refer to biovolume estimates for only those specimens for which M_E could be calculated as V_E-partial. Each seafloor image was also visually classified to habitat type: (i) hard substrata (boulder, cobble; total 54 m²), (ii) sand (1169 m²), (iii) mud (2034 m²), and (iv) mosaic where there was substantial hard substratum present in sand or mud (10–50% areal coverage; 618 m²; e.g. Benoist et al., 2019). To acknowledge the likely inhomogeneity of variance and the unbalanced sampling design, standing stock data (biovolume or biomass standardised to seafloor area observed) were $\log_{10}$-transformed and assessed using Welch's ANOVA (Welch, 1951) with subsequent pairwise comparisons made using the Games-Howell method (Games and Howell, 1976), as implemented in Minitab.

2.4.2. Inter-operator variation assessment

Two operators (O1, O2) were trained jointly to apply GVM body-size measurements on a selection of photographed individual mega-benthos specimens representative of the range of taxa encountered in the study area. Training consisted of repeat measurements of the selected specimens and joint quality/control assessment of the resultant data to minimise intra- and inter-operator inconsistencies. This process yielded two final training samples of 130 paired specimen records (V_E-training). Each operator then independently processed a set of c. 2400 images. A subset of 20% of those images was analysed by both operators (i.e. c. 480 images common to O1 and O2), yielding two field samples of estimated standing stock (M_E, V_E-partial, V_E). To measure variability in standing stock estimates between operators, the training and the field datasets were randomly resampled with replacement to establish an appropriate measure of inter-operator error in standing stock estimation using a modified form of bootstrapping (Davison, 1997). This process was repeated 10,000 times using a custom script implemented in the R environment (R Core Team, 2016). For each dataset, total standing stock (V_E-training, M_E, V_E-partial, V_E) was calculated for each bootstrap sample, and the 95% confidence interval (CI) of relative difference between operators was estimated using the simple percentile method (Davison, 1997). To give context to the inter-operator variability estimates, the overall relative variability in field standing stock was also estimated using the same method for the same sampling unit size (i.e. 10,000 resamples with replacement of c. 480 images from the complete image set).

3. Results and discussion

3.1. Evaluation of current methods

3.1.1. Trawl sample versus photographic survey

Fig. 3 presents the megabenthos biomass data (invertebrates and fish) from the Wei et al. (2010) dataset as divided into trawl catch and photographic survey methods. These data suggest substantial underestimation of biomass when based on trawl catches. More formally, the LM of log(biomass) on water depth by method reveals a statistically significant effect of method ($F_{1,220} = 76.4$, $p < 0.001$) and no
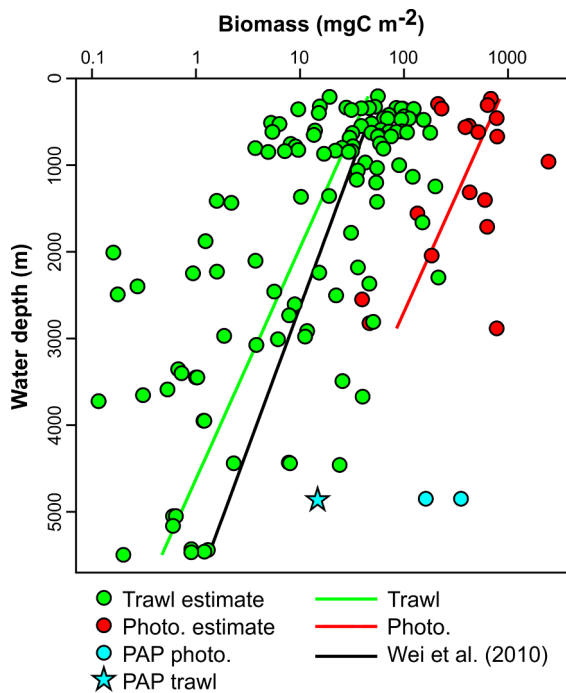
Fig. 3. Megabenthos biomass as a function of water depth and estimation method: trawl catches and photographic surveys. Data shown are as compiled by Wei et al. (2010) with the addition of (i) two values for the Porcupine Abyssal Plain Sustained Observatory site derived from recent photographic assessments (Durden et al., 2015; Morris et al., 2016), and (ii) the 'true' trawl estimate of PAP-SO megabenthos biomass (invertebrate only; Billett et al., 2001, 2010), with units of mass converted from fresh wet weight to carbon mass using the coefficients provided by Brey et al. (2010). Lines represent regressions of log(biomass) on water depth: (i) black, original regression provided by Wei et al. (2010), with general linear model results for (ii) green, trawl catches, and (iii) red, photographic surveys. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Fig. 4. Length-weight relationship of the holothurian *Psychropotes longicauda* sampled from the Porcupine Abyssal Plain Sustained Observatory between 2004 and 2017. Scatter plot of individual values with corresponding log–log regression (red line) and associated 95% prediction interval (blue lines); $F_{1,982} = 4232.4$, $p < 0.001$, $R^2 = 81.1\%$, $W = 0.442 \times SL^{2.42}$. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

statistically significant interaction between water depth and method ($F_{1,219} = 0.75$, $p = 0.388$). Consequently, the model predicts a common slope for the response of log(biomass) to water depth for both methods of -3.75×10^{-4}, with a 95% CI of -4.32×10^{-4} to -3.17×10^{-4} that does not encompass the slope originally published (i.e. -3.07×10^{-4}; Wei et al., 2010). In particular, note the substantial offset in the estimated intercepts: trawl 1.73 (95% CI 1.59, 1.88) and photo 3.01 (95% CI 2.87, 3.15). This equates to a factor of 20 underestimation of biomass at a given water depth by trawl catch relative to photographic survey.

We suggest that this substantial, systematic, methodological bias should be carefully considered, particularly when estimates of standing stock biomass are being produced (e.g. Wei et al., 2010), and where those estimates are being used in future climate change scenario forecasts (e.g. Jones et al., 2014). The current state of knowledge may be substantially biased towards underestimated stocks of seafloor biomass. Fig. 3 illustrates two additional biomass estimates from the PAP-SO site as provided by recent photographic assessments (Durden et al., 2015; Morris et al., 2016). The mean value of these estimates, 258 mgC m^{-2} (photographic data), is over two orders of magnitude greater than the corresponding estimate from the original regression of 2 mgC m^{-2} (pooled trawl and photographic data; Wei et al., 2010). The 'true' trawl estimate of PAP-SO megabenthos biomass (invertebrate only) is 17 mgC m^{-2}, based on 44 otter trawls covering the period 1989–2005 (Billett et al., 2001, 2010). We conclude that the state-of-the-art in megabenthos biomass estimation (e.g. Wei et al., 2010) is generally a factor of 20 below the likely true value and may be underestimated by a
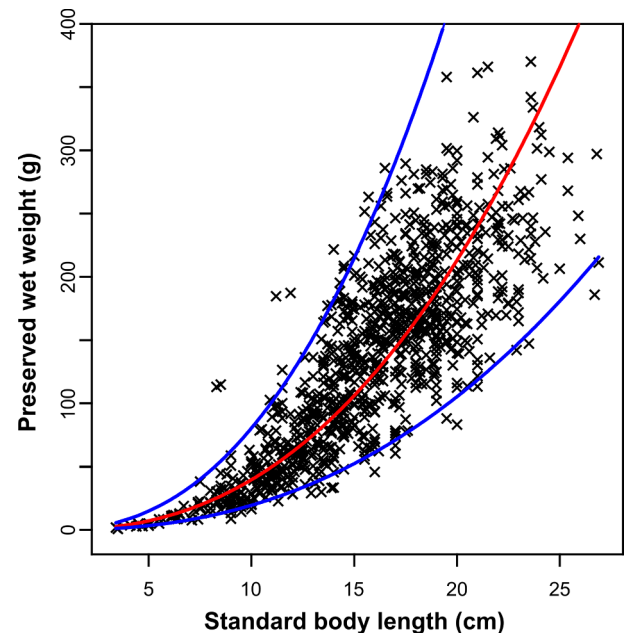
factor of 200+ locally. This is not a new observation, nor is the potential use of underwater photography to tackle the issue a new solution (e.g. McIntyre, 1956; Uzmann, 1977; Bett et al., 2001). We therefore suggest that there is both a clear need and scope for substantial improvement via the widespread adoption of photographic-survey-based megabenthos biomass estimation.

3.1.2. Evaluation of LWR method

Illustrated in its linear form, the LWR for *P. longicauda* demonstrates the rather diffuse nature of that relationship (Fig. 4). For example, a specimen of 20 cm standard length ($19.5 < SL \leq 20.5$ cm) has an observed weight range of 84–358 g preserved wet weight (pwwt) and a predicted weight range of 203–226 g pwwt using the LWR method. Although the LWR approach may superficially appear to be an exact numerical method, i.e. individual biomass is obtained mathematically from a standard measurement, it is effectively an approximate technique, with the predicted value ranging 40–170% of the true value in this example.

The LWR illustrated in Fig. 4 represents a composite of 15 trawl catches from seven cruises spanning almost 15 years. When the identities of individual catches, or cruises, are included in a LM, statistically significant variation is evident in both cases (trawls $F_{1,14} = 3.54$, $p < 0.001$; cruises $F_{1,6} = 4.97$, $p < 0.001$), and in both cases there are statistically significant interactions with standard body length (length × trawl $F_{1,14} = 2.86$, $p < 0.001$; length × cruise $F_{1,6} = 2.77$, $p = 0.011$). Consequently, there are statistically significant differences in both the *a* and the *b* parameters among trawls and cruises (e.g. Fig. 5). Such variation calls in to question the selection of the most appropriate value for these parameters in the case of *P. longicauda* or indeed any other species.

Fig. 6 illustrates the variation in the relative weight of *P. longicauda* by year of sampling. Spearman's rank correlation of relative weight and standard body length indicated the effective independence of these measures ($r_{s984} = -0.037$, $p = 0.247$). A Mood's median test detected statistically significant temporal variation in relative weight
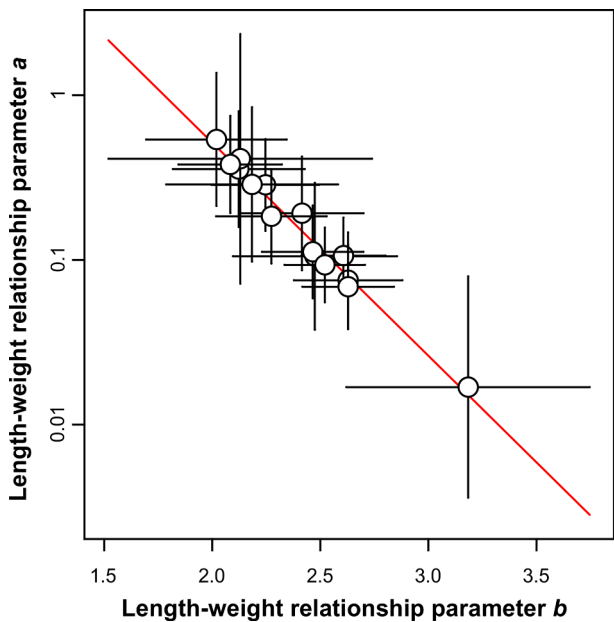
Fig. 5. Variation in length-weight relationship (LWR) of the holothurian *Psychropotes longicauda* sampled from the Porcupine Abyssal Plain Sustained Observatory between 2004 and 2017. For each trawl catch, the *a* and the *b* parameters of the LWR are illustrated, together with corresponding 95% confidence intervals. The red line represents the log-linear regression of *a* on *b*. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)
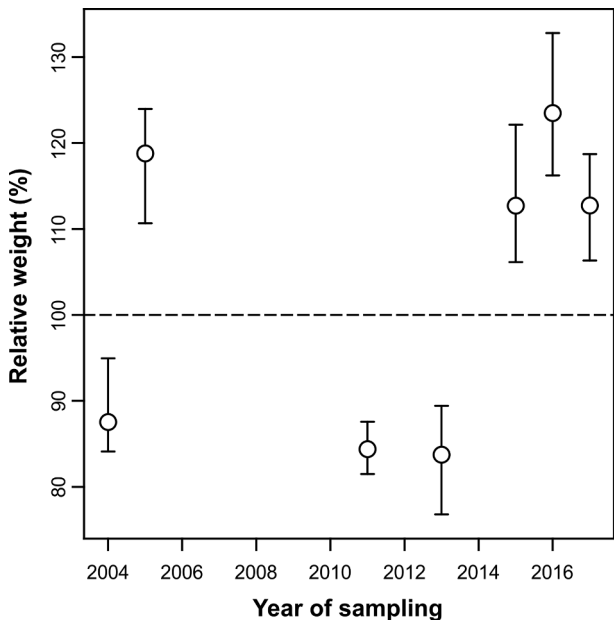

Fig. 6. Temporal variation in the relative weight (specimen weight/LWR-predicted weight; Eq. (2)) of the holothurian *Psychropotes longicauda* from the Porcupine Abyssal Plain Sustained Observatory between 2004 and 2017. The median value is presented with corresponding 95% confidence interval. Dashed line serves as reference line.

($\chi^2_6 = 136.8$, $p < 0.001$). In effect, specimens collected in 2005 and 2015–2017 were c. 15% heavier at a given length than the general LWR prediction, and those collected in 2004, 2011, and 2013 were c. 15% lighter than the prediction. Such systematic temporal variation could, for example, be linked to the known intra- and inter-annual variation in the supply of organic matter to the seafloor in this location (e.g. Bett et al., 2001). This, again, calls in to question the selection of the most

Table 1
Relationships between biomass and biovolume of megabenthos specimens from the Porcupine Abyssal Plain Sustained Observatory, as directly measured and photographically estimated using the generalised volumetric method and the length-weight-relationship approach. Pearson's product-moment correlation coefficients (r) of measured ($_M$) and estimated ($_E$) specimen mass (M_M, M_E; g fwwt) and volume (V_M, V_E; mL), for linear (lin.) and logarithmic (log.) relationships (in all cases $n = 206$, and $p < 0.001$).

Variable	M_M (lin., log.)	V_M (lin., log.)	M_E (lin., log.)
V_M (lin., log.)	0.997, 0.981		
M_E (lin., log.)	0.897, 0.977	0.901, 0.960	
V_E (lin., log.)	0.952, 0.934	0.954, 0.923	0.907, 0.941

appropriate LWR parameter values for *P. longicauda* or any other species.

3.2. Validation with physical specimens

We tested the relationships between direct physical measurement (M_M, V_M) and indirect photographic body-size measurement (M_E, V_E) obtained from a variety of trawl-caught specimens from the PAP-SO study site (Appendix A). All of the relationships examined between measured and estimated mass and volume yielded strong and statistically significant Pearson's product-moment correlations (r), ranging between 0.897 for $M_M \sim M_E$ to 0.997 for $M_M \sim V_M$ (Table 1). Consequently, there was only minor variation between the regression slope coefficients estimated by the model I (Table 2) and the model II (Table 3) methods. Indeed, in most cases the 95% CI of the regression slope encompasses the value 1.0; the primary exception being the relationship $M_M \sim M_E$ that yielded substantially lower slope values (c. 0.6; Tables 2 and 3).

Similarly, ordinary least squares regressions were all statistically significant and exhibited good predictive capacity with predicted R^2 ranging between 76.1% for $M_M \sim M_E$ to 99.4% for $M_M \sim V_M$ (Table 2). All OLS regression coefficients were statistically significant, except in the case of the intercept for $M_M \sim V_M$ in both linear and logarithmic forms, suggesting a very close correspondence between body mass and body volume (Table 2). In other words, for every additional mL in body volume, body mass is expected to increase by c. 1 g fwwt, suggesting an average tissue volumetric mass density of 1.053 (linear) and 1.058 (logarithmic; Fig. 7a; Table 2), with very similar values derived from the linear forms of RMA (1.056) and SMA (1.056) model II regressions (Table 3).

Among the fresh specimens, the proposed GVM appeared to have good predictive capability for both volume (predicted R^2 89.4–92.0%; Fig. 7c) and mass (89.7–95.4%; Fig. 7b), which exceeded that of mass prediction by the conventional LWR method (76.1–86.9%; Table 2). In practical terms, the standard deviation of the estimate was 47.1 g fwwt for the GVM and 68.1 g fwwt for the LWR method, where the corresponding value from directly measured volume was 11.6 g fwwt.

Although the estimation of mass from standard length is a commonly applied technique, it is not entirely surprising that the proposed GVM has an improved predictive capacity. The estimation of mass from a single measured dimension (SL) relies on a consistent relationship between the measured dimension and the two unmeasured dimensions, whereas the volumetric approach measures two dimensions, with the second-dimension measurement (ECD) subjectively modified to be representative of the third unmeasured dimension. Further, as previously discussed, the mass of an individual of a given standard length may be, for example, substantially influenced by its life stage, physical condition, feeding success, health, season, and geographic location (e.g. Kimmerer et al., 2005; Méthot et al., 2012; Meyer, 1989; Primavera et al., 1998; Zilli et al., 2017).

The volumetric assessment of individual biomass is frequently used in the study of microscopic and small-bodied organisms (Baguley et al.,

Table 2
Predictive capacity of photographic methods to estimating body size. Model I linear regressions between measured and photographically estimated body size of megabenthos specimens from the Porcupine Abyssal Plain Sustained Observatory, using the generalised volumetric method and the length-weight-relationship approach. Results of model I ordinary least squares regression analyses of measured ($_M$) and estimated ($_E$) specimen mass (M_M, M_E; g fwwt) and volume (V_M, V_E; mL). (Pred., predicted; CI, confidence interval; ***, $p < 0.001$).

Equation	$F_{[1,204]}$	R^2 (%)	Pred. R^2 (%)	Intercept 95% CI	$t_{[204]}$	Slope 95% CI	$t_{[204]}$
$M_M = 1.250 + 1.053\ V_M$	35768***	99.4	99.4	(-0.543, 3.043)	1.37	(1.042, 1.064)	189***
$\ln(M_M) = 0.056 + 0.996\ \ln(V_M)$	5254***	96.3	96.2	(-0.036, 0.147)	1.20	(0.968, 1.023)	72.5***
$M_M = 13.91 + 0.982\ V_E$	1992***	90.7	89.7	(6.831, 20.99)	3.87***	(0.939, 1.026)	44.6***
$\ln(M_M) = 0.472 + 0.938\ \ln(V_E)$	4303***	95.5	95.4	(0.381, 0.562)	10.3***	(0.910, 0.966)	65.6***
$M_M = 20.49 + 0.547\ M_E$	844***	80.5	76.1	(10.32, 30.67)	3.97***	(0.510, 0.584)	29.1***
$\ln(M_M) = 0.408 + 0.837\ \ln(M_E)$	1386***	87.2	86.9	(0.248, 0.568)	5.03***	(0.793, 0.882)	37.2***
$V_M = 12.11 + 0.931\ V_E$	2057***	91.0	89.4	(5.500, 18.72)	3.61***	(0.891, 0.972)	45.4***
$\ln(V_M) = 0.504 + 0.908\ \ln(V_E)$	2403***	92.2	92.0	(0.387, 0.622)	8.48***	(0.872, 0.945)	49.0***

2004; Mustard and Anderson, 2005; Novack-Gottshall, 2008; Mazurkiewicz et al., 2016). Studies of macro- and megafaunal marine organism biovolume have previously been used as an indicator of relative biomass, or the physical space occupied by individuals (McClain, 2004, 2009; Jones et al., 2007). In the present study, we have been able to compare directly mass estimates by our proposed taxon-independent GVM and the taxon-specific LWR method, where those relationships were derived from a very extensive measurement database of c. 47,000 specimens (Durden et al., 2016a). The GVM appears to outperform the traditional LWR method, having a higher predicted R^2 value and lower standard error of estimate value. The generalised volumetric estimate (V_E) does requires the user to measure two body dimensions (*ECD*, *ECL*). Nonetheless, the additional time (cost) of making the second measurement is small compared to the full process of locating a specimen within an image, identifying that specimen, and making a single measurement (*SL*). The proposed method is taxon, time, and location independent, offering considerable benefits that are further explored in the following case study.

3.3. Celtic Sea case study trial

In total, 2896 specimens from eight phyla and 92 taxa were measured from photographs using both the GVM and the LWR approach (Appendix A). The estimated range for individual biovolume (V_E) was 0.001 mL to 16.98 L, and for biomass (M_E) 0.001 g to 17.35 kg. Total standing stock estimated by the GVM was very similar to that estimated with the LWR method (V_E-*partial* 7.74×10^{-3} mL m^{-2}, M_E 7.34×10^{-3} g m^{-2}). No statistically significant differences were detected between V_E-*partial* and M_E estimates for the total surveyed area, or within the individual habitat types encompassed by the survey (Fig. 8). Similarly, both methods illustrated the same pattern and detected the same statistically significant differences between habitat types (V_E-*partial* $F_{3,267} = 46.69$, $p < 0.001$; M_E $F_{3,266} = 53.13$, $p < 0.001$). The same pattern and statistically significant differences were also apparent in the total biovolume data, i.e. including those taxa

for which LWR estimation was not possible (V_E $F_{3,274} = 131.67$, $p < 0.001$). However, there were appreciable increases in estimated standing stock from the M_E and V_E-*partial* values to the V_E values (Fig. 8). By application to the full range of taxa present, the GVM increased the total standing stock estimate over the LWR method by a factor of 1.6 for the total seafloor area surveyed, by about double in the case of mosaic and mud habitats, and around four-fold on hard substrata.

The potential advantage of the GVM, compared to the LWR method, was well demonstrated in the Celtic Sea case study trial. This region encompasses substantial areas of mixed substratum types (mosaics of hard rock and mobile sediments) that are not easily surveyed using physical sampling methods, such that photography may be the only uniformly applicable approach to stock assessment across habitat types. Estimated biovolume (V_E-*partial*) was highly consistent with the biomass estimates (M_E) obtained by the LWR method, suggesting at least an equal performance for the proposed method. Further, the volumetric method enabled the assessment of the c. 25% of taxa for which no LWR data were available (mainly bryozoans, sponges, and colonial cnidarians). Located in the European Atlantic shelf seas, the fauna of this study area is very well known with a substantial literature from which to derive LWR conversion factors (Coull et al., 1989; Robinson et al., 2010; Silva et al., 2013; Benoist et al., 2019). However, in marine regions lacking that information, the proposed taxon-independent GVM offers the prospect of useful standing stock assessments despite a lack of taxon-specific information. In addition, the volumetric approach enables the assessment of those organisms that do not exhibit a distinctive body form or that are rarely sampled as complete entities (e.g. sponges, colonial and encrusting taxa).

The Celtic Sea dataset was produced by two different operators trained to apply GVM body-size measurements using a common training image dataset. Following that training, there was no statistically significant difference in the total volume estimated (V_E-*training*) between operators O1 and O2 (Fig. 9a). This preliminary test was further expanded in the full field trial. As suggested by Durden et al.

Table 3
Functional relationship between photographically estimated and measured body size. Model II linear regression between measured and photographically estimated body size of megabenthos specimens from the Porcupine Abyssal Plain Sustained Observatory, using the generalised volumetric method and the length-weight-relationship approach. Results of model II regression (RMA, ranged major axis; SMA, standardised major axis) analyses of measured ($_M$) and estimated ($_E$) test specimen mass (M_M, M_E; g fwwt) and volume (V_M, V_E; mL). (CI, confidence interval).

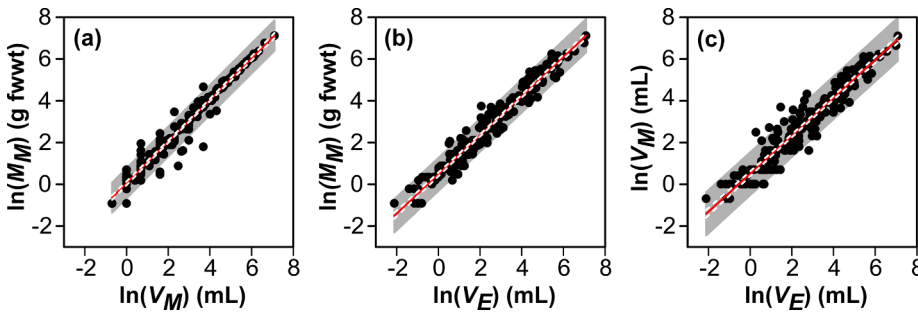| Equation	RMA intercept (95% CI)		RMA slope (95% CI)		SMA intercept (95% CI)		SMA slope (95% CI)	
$M_M \sim V_M$	1.019	(0.199, 1.830)	1.056	(1.045, 1.067)	1.028	(-0.766, 2.822)	1.056	(1.045, 1.067)
$\ln(M_M) \sim \ln(V_M)$	0.002	(-0.077, 0.079)	1.014	(0.987, 1.042)	0.001	(-0.039, 0.040)	1.015	(0.988, 1.042)
$M_M \sim V_E$	10.62	(7.517, 13.58)	1.032	(0.987, 1.078)	10.65	(3.497, 17.80)	1.031	(0.989, 1.076)
$\ln(M_M) \sim \ln(V_E)$	0.410	(0.335, 0.483)	0.962	(0.934, 0.992)	0.180	(0.141, 0.220)	0.960	(0.932, 0.989)
$M_M \sim M_E$	13.34	(8.703, 17.66)	0.614	(0.573, 0.657)	13.78	(3.378, 24.18)	0.610	(0.574, 0.648)
$\ln(M_M) \sim \ln(M_E)$	0.226	(0.082, 0.362)	0.899	(0.853, 0.948)	0.101	(0.031, 0.171)	0.897	(0.853, 0.942)
$V_M \sim V_E$	9.210	(6.330, 11.97)	0.975	(0.933, 1.018)	9.109	(2.437, 15.78)	0.977	(0.937, 1.018)
$\ln(V_M) \sim \ln(V_E)$	0.397	(0.297, 0.492)	0.950	(0.913, 0.990)	0.177	(0.126, 0.228)	0.946	(0.910, 0.983)

Fig. 7. Comparison of measurements and estimates of volume and biomass of fresh megabenthos specimens from the Porcupine Abyssal Plain Sustained Observatory. Log-log linear regressions. (a) Measured mass (M_M) on measured volume (V_M). (b) Measured mass (M_M) on geometrically estimated biovolume (V_E; Eq. (3)). (c) Measured biovolume (V_M) on geometrically estimated biovolume (V_E). Solid red lines are regressions, dashed white lines are corresponding 95% confidence intervals, and shaded areas the corresponding 95% prediction intervals.
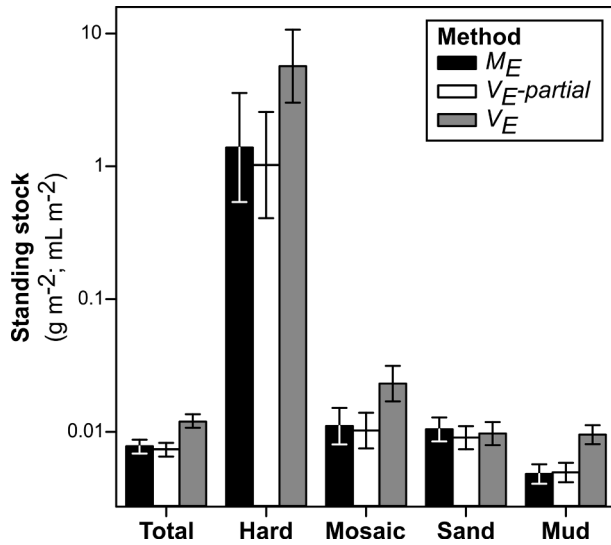
3.4. Generalised volumetric method

Individual body size and total biomass of the megabenthos are essential variables given their central roles in the regulation of marine ecosystems. Yet, the lack of an appropriate and cost-effective method, applicable at broad scale, has limited their consideration in the Framework for Ocean Observing. The increasing use of robotic technologies, remotely operated vehicles and particularly autonomous underwater vehicles, to study the seafloor, has delivered new opportunities for the quantitative assessment of the megabenthos across a range of spatial scales and environments. The collection and analysis of large amounts of photographic data (digital stills, digital video, chemical films) does bring new challenges, including the estimation of biomass in the absence of physical specimens or prior knowledge of LWRs. In these cases, we suggest that the use of the generalised volumetric method (GVM) offers an effective means to estimate biovolume. Indeed, given that the volumetric method appears to outperform the LWR method even where extensive prior information is available (e.g. PAP-SO and UK Celtic Sea sites), we would suggest that it is considered for use more generally. We would also again note that there is clear evidence that LWRs can exhibit substantial taxon, time, and location, specificities that have the potential to introduce appreciable biases to biomass estimates where those variations in LWRs are not known or controlled. It may also be worth noting that to describe how individuals acquire and use energy, some ecological models adopt biovolume as their main body-size currency, such as dynamic energy budget (DEB) models (Kooijman, 2000) that typically include 'structural length' (i.e. biovolume$^{1/3}$) as a primary variable (e.g. Sousa et al., 2010), under the assumption that most physiological processes are volume dependant.

The GVM does require the user to convert virtually the specimen into a compressed cylinder of equivalent volume in order to best estimate *ECD* and *ECL*. We would therefore recommend the user should have significant zoological experience, be familiar with the morphology of the taxa involved, and ideally, have prior experience with handling comparable physical specimens (see recommendations in Section 2.2).
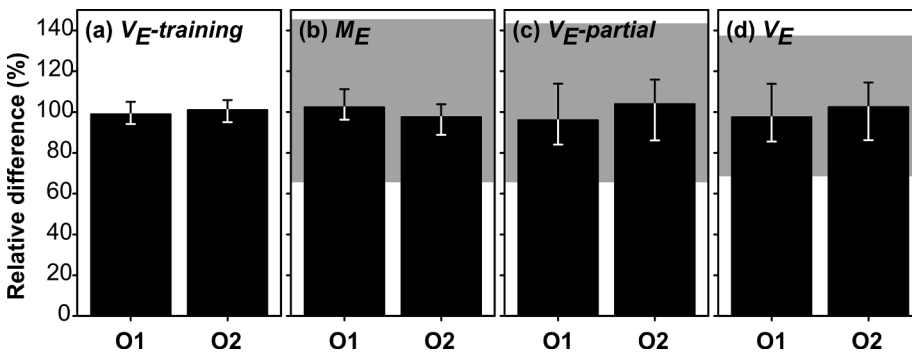

Fig. 8. Celtic Sea megabenthos standing stock biomass by habitat type and estimation method. The mean value is presented with 95% confidence interval, as estimated using the length-weight-relationship (LWR) approach (M_E, g m^{-2}; Eq. (1)) and the generalised volumetric method (mL m^{-2}; Eq. (3)), excluding (V_E-partial) and including (V_E) those taxa for which LWR estimation was not possible.

(2016b), a subset of images were analysed by both operators to quantify potential inter-operator bias in the estimation of mass and volume using both the LWR and the GVM approaches. There were no statistically significant differences in estimated biomass (M_E) or estimated biovolume (V_E-partial, V_E) between operators (Fig. 9b–d). Note also that the field sample relative variability was appreciably greater than the inter-operator relative variability for both LWR and GVM approaches (Fig. 9b–d).

Fig. 9. Inter-operator variability in the estimation of Celtic Sea megabenthos standing stock biomass. Variability, as 95% confidence interval of individual operator mean value, is illustrated as relative difference (%) from the joint mean value (i.e. 100%) of the two operators (O1, O2). (a) Total biovolume estimated by the generalised volumetric method (GVM) for an initial training dataset (V_E-training). (b) Total survey biomass estimated using the length-weight-relationship (LWR) approach (M_E, i.e. Eq. (1)). (c–d) Total survey biovolume estimated using the GVM, excluding (c; V_E-partial, i.e. Eq. (3)) and including (d; V_E, mL) those taxa for which LWR estimation was not possible. The shaded bands (b–d) represent the variability, as 95% confidence intervals, of the full survey estimates of the corresponding standing stock parameter (i.e. M_E, V_E-partial, V_E).

The GVM necessarily incorporates a subjective element in this conceptual specimen-to-cylinder conversion. Consequently, multiple users contributing to a common dataset should inter-calibrate their approach, as we have illustrated in the Celtic Sea case study trial. It may be impossible to eliminate all such differences in measurement within and between analyses (e.g. Mazurkiewicz et al., 2016); however, some simple precautions, such as randomisation, can readily be implemented (see recommendations in Durden et al., 2016b, c). For example, in the Celtic Sea case study trial, we ensured that each operator was randomly allocated images from the full image set available to avoid bias between AUV deployments and between habitat types. Similarly, we randomised the order in which each operator assessed images in order to avoid temporal variation in the operator's performance being unintentionally translated into spatial variation, had the images been analysed in the original field sequence.

There are clearly opportunities for further development of the generalised volumetric method. One is in the automation of the basic process, as has been achieved for particulate organic matter (Iversen et al., 2010) and nematode biovolume assessment (Moore et al., 2013; Mazurkiewicz et al., 2016). This could be more challenging in the case of megabenthos in seafloor photographs; firstly, because the complex background (i.e. the seafloor) makes in situ specimen delineation more involved, and secondly, because of the wide variety of body shapes exhibited across taxa. Nevertheless, automation could be achieved through recent rapid advances in machine vision and in machine learning (Schoening et al., 2012, 2016; Langenkämper and Nattkemper, 2017). Machine recognition of basic morphological types could enable automated application of our proposed method. A second challenge will be to improve the conversion of estimated biovolume (fresh wet weight mass) to units of carbon mass and energy that may be particularly valuable in the application of numerical modelling frameworks such as the metabolic theory of ecology (Brown et al., 2004) and DEB (Kooijman, 2000) models in the assessment of ecosystem stocks and flows. Conversions from wet weight are widely available (e.g. Brey et al., 2010) and serve as a useful approximation, i.e. by assumption of volumetric mass density (e.g. 1.056; see Section 3.2).

4. Conclusions

Biomass is a key ecological variable that informs the fields of conservation, environmental quality assessment, resource management, and the study of the stocks and flows of mass and energy through ecosystems. It is featured as an essential biodiversity variable (EBV) and as an 'emerging' essential ocean variable (EOV), prompting the need for a method for the measurement of individual biomass, which is broadly applicable and which can be readily adopted by a wide range of users. In seafloor imagery, the traditional LWR approach employed to derive individual biomass relies on pre-existing taxon-specific data and may be subject to systematic, temporal and spatial, variation. The LWR method is also restricted to readily sampled taxa that have a fixed body form. These significant limitations may be overcome with the taxon-independent generalised volumetric method described here. The predictive ability of the GVM, in accuracy and in precision, appears to at least equal that of the LWR approach, and it has much more general and much more immediate applicability.

Declaration of Competing Interest

None.

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Appendix A. Supplementary material

Supplementary data to this article can be found online at https://doi.org/10.1016/j.pocean.2019.102188.

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