

Efficiently measuring complex sessile epibenthic organisms using a novel photogrammetric technique

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Abstract

This paper describes a stereo photogrammetry method that allows accurate measurements of volume for sessile epibenthic organisms. It represents a novel approach based on multiple views (five stereo image pairs) from a purpose built stereo digital still camera system combined with a three-dimensional reconstruction software program (CAM). The bias, accuracy, precision and efficiency of the method were assessed in the laboratory using models with three levels of morphological complexity (simple, moderate and complex morphologies) and two sizes (large and small). The technique did not show any biases to observer experience, with no significant difference among observers ($p > 0.05$). Volume measurements made with CAM were very accurate when compared with the water displacement volume of each model, with an overall mean error of about -3% (S.E. $\pm 1\%$). The CAM volume measurements were more accurate on complex models and moderately complex models than on models with simple morphologies. Also, large models had a higher accuracy than small models. Volume measurements made with CAM were also highly precise with the lowest precision observed being $\pm 2\%$ of the volume estimate. The time required for a volume estimation using the CAM method was also highly efficient, and the longest time taken for a volume estimation was on average 1 h and 36 min, making it the fastest reported three-dimensional reconstruction method. In field applications, volume estimations of four sponges were all within the observed accuracy, precision and efficiency established during laboratory trials. The accuracy, precision and efficiency demonstrated by the CAM method make this technique highly suitable for routine measurements of the volume of sessile epibenthic organisms.

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1. Introduction

Measurements of volume, biomass or surface area are particularly important when determining patterns of growth and secondary production by an organism or benthic community (Edgar, 1990; Kingsford and Battershill, 1998; Eleftheriou and McIntyre, 2005).

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Sponges, like many benthic invertebrates, exhibit a bewildering array of morphologies (Jackson, 1979; Bell and Barnes, 2000) that are highly plastic (Palumbi, 1984, 1986; Kaandorp, 1999). This makes obtaining repeatable and accurate measurements of size or biomass difficult.

The biomass and size of sponges have been measured using a variety of techniques including simple morphometric measurements (Reiswig, 1973; Duckworth and Battershill, 2001, 2003a; McDonald et al., 2002, 2003; Duckworth, 2003), measurements of weight (wet or ash-free) (Barthel, 1986, 1988; Duckworth et al., 1997, 1999, 2004; Duckworth and Battershill, 2003b), as well as traditional measurements of surface area (Elvin, 1976) and volume (Wilkinson and Vacelet, 1979).

Many of these methods are destructive, like determining wet/dry weight or displacement volume as well as time consuming and can often produce highly variable results due to the loss of water from tissue (Duckworth et al., 1997). The use of destructive methods limits the type of research that can be conducted, particularly on rare species or when changes in the volume of an organism over time are of interest.

The problem with the use of non-destructive techniques such as simple morphometric measurements is that they are not very repeatable or accurate, particularly for morphologically complex animals such as sponges (Duckworth and Battershill, 2001). This limited ability to detect changes in the growth or biomass of epibenthic invertebrates impacts our ability to make inferences about the ecological processes involved (Duckworth and Battershill, 2001).

The size, shape and biomass of sessile organisms have recently been measured through photography and photogrammetry (Done, 1981; Ayling, 1983; Ben-Zion et al., 1991; Becerro et al., 1994; Harvey and Shortis, 1996; Ewins and Pilgrim, 1997; Leys and Lauzon, 1998; Shortis et al., 2000; Bythell et al., 2001; Deng and Faig, 2001; Duckworth and Battershill, 2001; Cocito et al., 2003; Duckworth, 2003; Handley et al., 2003; Eleftheriou and McIntyre, 2005). These methods vary from single camera photographic techniques (Ben-Zion et al., 1991; Leys and Lauzon, 1998; Duckworth, 2003; Handley et al., 2003), to methods of three-dimensional measurement and reconstruction using photogrammetry (Done, 1981; Shortis et al., 2000; Bythell et al., 2001; Cocito et al., 2003; Eleftheriou and McIntyre, 2005). The latter methods allow more accurate and repeatable measurements of size, shape and biomass without the destruction of the target organisms.

Photogrammetry has the advantage of being non-destructive, repeatable, and provides a permanent record

of the individual organism (Harvey and Shortis, 1996; Shortis et al., 2000). Additionally, stereo-photogrammetric techniques are not as limited as other methods by the size or orientation of the organism (Harvey and Shortis, 1996; Shortis et al., 2000). The disadvantages of stereo-photogrammetric techniques include costs associated with the equipment, clarity of images due to suspended materials, and time associated with processing images and time consuming measurements (Harvey and Shortis, 1996; Shortis et al., 2000). Problems with synchronisation of cameras can also cause problems with measurements (Eleftheriou and McIntyre, 2005).

To date, stereo-photogrammetry has not been used to measure sponges, with most uses in marine research involving fishes (Harvey et al., 2001a,b, 2002, 2003), dolphins (Braeger et al., 1999), shellfish (Shortis et al., 2000) and hard corals (Done, 1981). However, a few studies have utilised single camera photogrammetry to measure the morphology of other benthic organisms such as hard corals (Bythell et al., 2001; Cocito et al., 2003).

Recently, software and imaging technology developments have facilitated accurate and quantitative measurements of benthic organisms. Bythell et al. (2001) and Cocito et al. (2003) used underwater photogrammetry to make three-dimensional measurements of hard corals. Both studies report accurate morphological measurements from their respective techniques, but both were constrained by equipment and the need for scale and reference objects (Bythell et al., 2001; Cocito et al., 2003). This limits these methods when researching organisms in confined or overhanging environments, as is typically the case with sponges. The techniques were also restricted by long data processing/analysis times, with moderate image quality and resolution that restricted measurement accuracy (Bythell et al., 2001; Cocito et al., 2003). In addition, both techniques also relied on the combination of separate single images for the photogrammetric analysis. The robustness of these techniques was reduced as the slightest movement of the subject during image acquisition could introduce errors in the measurement. This limited the application of these techniques in studying delicate/erect sponge species which would have some movement with water turbulence.

A stereo-photogrammetric method for obtaining accurate measurements of complex biological objects needs to:

1. Be able to work in restrictive spaces.
2. Be able to investigate relatively large areas, combining the ability to research one organism or numerous organisms.

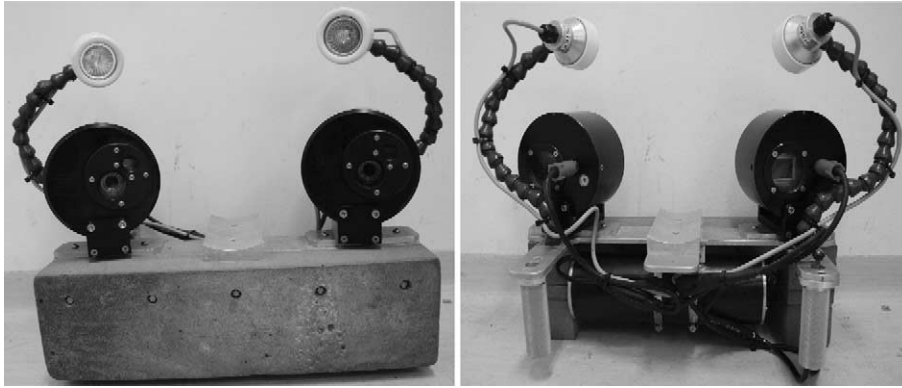


Fig. 1. Photograph of stereo camera system used in this study. The left image shows the front of the stereo system and the right image shows the rear of the stereo system.

3. The acquisition of data must be uncomplicated, easily performed *in situ* and must be time efficient.
4. The process of obtaining a measurement must be easy to perform, precise, accurate and completed in a practical amount of time.

To overcome the limitations of current photogrammetry techniques, and those destructive traditional techniques we developed a photogrammetric technique for measuring the volume of sessile epibenthic invertebrates using stereo-photogrammetry. The aims of the research were to:

1. Examine the minimum number of stereo image pairs required to make a volume measurement.
2. Evaluate the observer bias, accuracy, precision and efficiency of the technique under laboratory trials.
3. Investigate the effectiveness of the technique under field conditions.

2. Materials and methods

2.1. Camera setup

All images were captured using a stereo digital still camera system (stereo system) (Fig. 1). The stereo system consists of two Canon™ Powershot S45 four mega-pixel digital still cameras contained within aluminium underwater housings. The camera housings were mounted onto a solid base bar 336 mm apart, with the cameras being angled slightly towards each other (15 degrees), resulting in an overlapping Field of View (FOV) (Fig. 2). The stereo system was also equipped with twin 50W halogen video lights connected to a Ni-mH power pack mounted underneath the camera base bar for low light conditions.

The cameras were manually set for a fixed focal length (50 cm), aperture (F4.0), shutter speed (1/100 s) and ISO (ISO 200) to ensure that the zoom, focus or aperture did not change during image acquisition. Any changes in these parameters can severely affect camera calibration and the accuracy of the resulting photogrammetric measurements (Harvey and Shortis, 1998).

2.2. Camera calibration

To make accurate three-dimensional measurements the stereo system must first be calibrated as described by Harvey and Shortis (1996). Calibration is achieved by capturing images of a calibration cube in various orientations. The images are imported into the computer

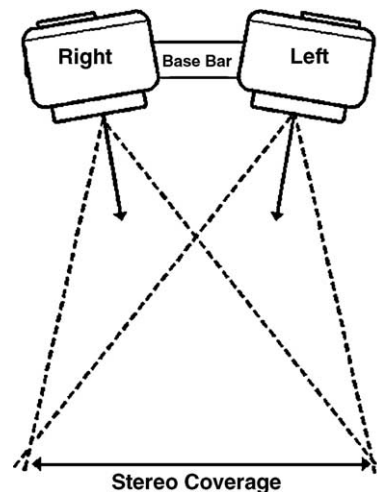


Fig. 2. Illustration of system setup to produce overlapping camera fields of view.

program Video Measurement System (VMS) (Robson and Shortis, 1995). This software estimates the optical and distortion properties of the cameras, as well as determines the relative orientations of the cameras. Calibration images were taken any time the cameras were removed from their housings as this may change the relative orientations of the cameras, and particularly the distance between the lens and port of the camera housing.

2.3. Measurement process

2.3.1. Software

Accurate morphometric measurements require a three-dimensional reconstruction of the target object. The three-dimensional reconstruction requires multiple stereo images to be recorded around the entire object as it is usually not visible from any single stereo image (Fig. 3). This was achieved by using a software program called Calibration and Measuring (CAM) developed by J. Seager.

2.3.2. Three-dimensional reconstruction

Reconstruction of the target object is achieved in CAM by creating a point cloud describing visible parts of the object in each stereo image pair. Once a sufficiently dense point cloud has been measured, object reconstruction including estimates of volume and surface area can be computed (note other quantities such as length, width and height can also be measured). Part of the modelling process requires the user to measure reference points to allow transformation of point coordinates into a single datum. The reference points can be natural (such as distinct features on or around the object being measured), or can be introduced via a reference object, usually in the form of bright dots on a dark background. These points do not need to be calibrated or have known positions relative to each other but need to be visible in each stereo pair so they can be measured.

CAM's algorithm generates a Delaunay triangulation (Guibas et al., 1992) using the user defined point cloud of the target object. However, for each camera location there

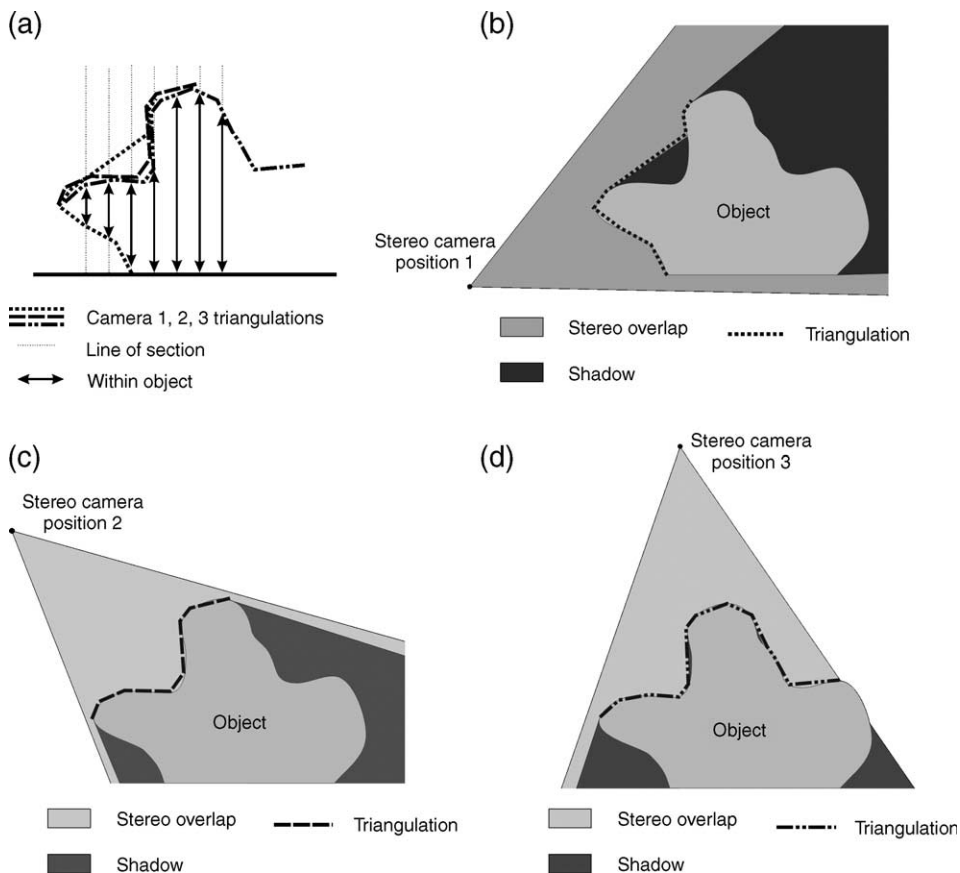


Fig. 3. Surface triangulation and cross-section generation. (a) Shows the construction of an accurate cross-section from triangulations in (b), (c), and (d), none of which is complete or accurate in its own right. Dashed lines represent each sample through the triangulation, and hard lines with arrowheads represent the matching 'into' and 'out of' object faces.

are parts of the object that are not visible. The triangulation from each camera position does not cover the entire surface of the object, and in some instances (Fig. 3(b–d)), it does not accurately describe the surface of the object due to shadowing. In this case the algorithm combines the triangulations from each set of stereo images to form a mass of overlapping triangulation meshes. CAM transforms the triangulations from all stereo images into the same coordinate system of the chosen reference stereo image pair. Three-dimensional model generation continues by forming a series of cross-sections through the overlaid triangulation meshes (Fig. 3a). The total volume and surface area of the object are calculated from the cross-sectional areas and perimeters by application of Simpson's Rule for areas and volumes (Bannister and Raymond, 1984; Whyte, 1984). The volume and surface area generation algorithm is automatic and results are generated graphically (Fig. 4) and in a summary file.

2.4. Laboratory trials

2.4.1. Model construction

Six models (three large models approximately 2.5 L in volume and three small models approximately 0.5 L in volume) were constructed using plasticine. The volume of each model was accurately determined in the laboratory

by measuring their water displacement (Eleftheriou and McIntyre, 2005). Simple (smooth surface with minor imperfections), moderately complex (surface had small to moderate bumps) and a highly complex (surface had large bumps with projections and concavities) model were constructed for both sizes (Fig. 5).

2.4.2. Number of images

To determine the minimum number of stereo image pairs required to estimate volume (\hat{V}), an increasing number of image pairs were used. Initially the overhead image pair was used and additional side view images (one at a time) were added until nine image pairs had been used. The minimum number of image pairs determined from this process was then used for all subsequent volume measurements.

2.4.2.1. Analysis. Using a two sample *t*-test significant differences ($p < 0.05$) between the \hat{V} from different combinations of stereo image pairs were compared for each model separately. Data were checked for normality prior to applying the *t*-test using an Anderson–Darling Test.

2.4.3. Technique evaluation

To evaluate the CAM technique four parameters were examined: observer bias, accuracy, precision and

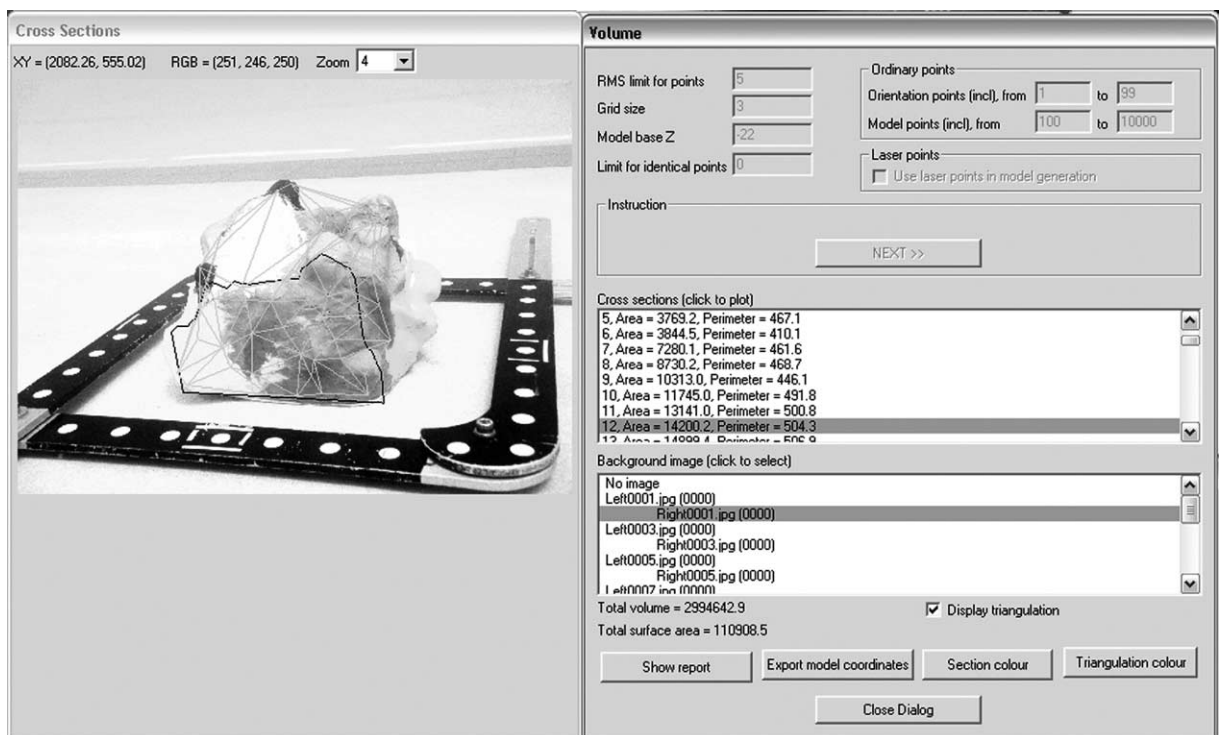


Fig. 4. Screen capture of CAM software triangulation and volume dialogue.



Fig. 5. Models used to evaluate the volume measuring technique. The large (left) and small (right) complexity models are the top images, moderate large (left) and small (right) complexity models are the middle images, and the large (left) and small (right) simple complexity models are the bottom images.

efficiency. Observer bias refers to any systematic deviation of \hat{V} for a model from its true volume (V) caused by observer artefacts; accuracy refers to how close a CAM \hat{V} is to the V of an object/organism (Zar, 1999); precision refers to the variability around \hat{V} (which may or may not be accurate) (Zar, 1999); and efficiency is how quickly \hat{V} can be determined for best accuracy and precision. Technique evaluation involved taking stereo images, completing three-dimensional reconstructions

and calculating \hat{V} for the six plasticine models. To test this technique underwater all imagery was captured in a swimming pool with a reference frame surrounding each model. Nine pairs of stereo images were taken, with volume estimations being made using the minimum number of stereo image pairs required (as determined above). These images and the necessary camera calibration data (obtained from VMS) were loaded into CAM, a three-dimensional reconstruction of the

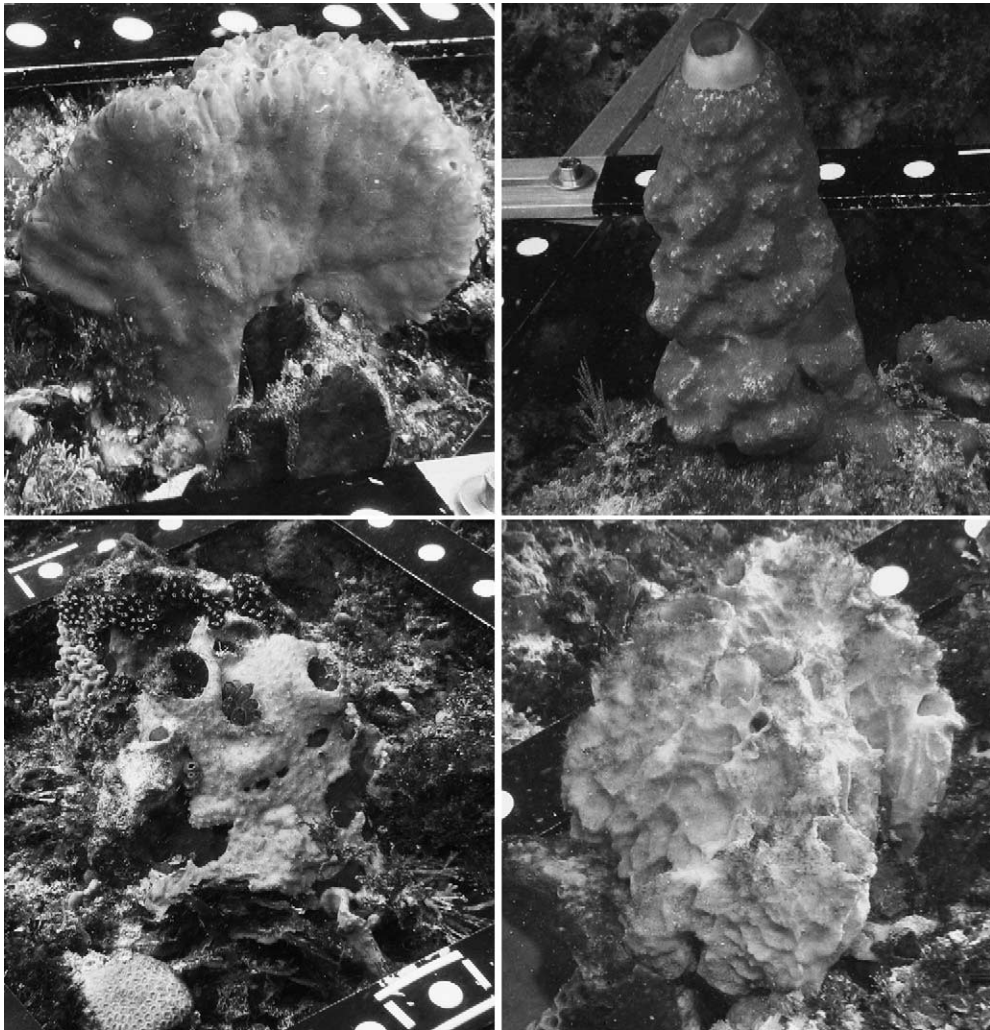


Fig. 6. Sponges sampled to test CAM in a field situation. Clockwise from top left is sponge 1 which has a flabellate morphology, sponge 2 (top right) has a tubular morphology, sponge 3 (bottom left) and sponge 4 (bottom right) both have massive morphologies.

object was produced and the \hat{V} of each model was determined.

2.4.3.1. Observer bias. To examine observer bias, three different novice observers calculated \hat{V} for each model three times. Increasing congruence among observers (represented by low variability among observers' \hat{V} for a model) results in a more accurate and precise \hat{V} for an object.

2.4.3.1.1. Analysis. After checking \hat{V} data for normality (Anderson–Darling Test) and equality of variance (Cochran's Test), observer biases were determined by comparing the three \hat{V} from each observer for each model using one-way ANOVA's.

2.4.3.2. Accuracy. Accuracy was calculated as the ratio between \hat{V} to V , where values greater than one correspond

to overestimations and values less than one correspond to underestimations.

$$\mathcal{A} = \hat{V}_x / V_x$$

Where: \mathcal{A} =accuracy, \hat{V} =estimated volume of model x , and V =true volume of model x .

Greater accuracy is achieved as the ratio of \hat{V}/V approaches one. This ratio also reveals any biases associated with the \hat{V} made by the stereo system (but not observer bias).

2.4.3.3. Precision. High precision is represented by low variability around the mean volume estimate for an object. The higher the precision, the greater power the CAM method has in making a correct volume estimate.

Precision is a function of the standard error of the sample, and has been described by several statistics but is appropriately and more commonly measured by the standard error (SE) to mean (\bar{x}) ratios (Andrew and Mapstone, 1987), the measure of precision adopted here.

$$\varphi = SE/\bar{x}$$

Where: φ = precision, SE = standard error (of the mean) of \hat{V} for model x , and \bar{x} = mean of \hat{V} for model x .

Greater precision occurs when the SE is small relative to the mean and precision decreases as the ratio increases.

2.4.3.4. *Efficiency.* The time taken to complete a three-dimensional reconstruction, as well as the time

to complete other tasks (for example image download and calibration) to make a \hat{V} was recorded for each model. This allowed an assessment of the overall time cost of the technique, as well as determining the minimum investment for greatest accuracy for each model.

2.5. Field trials

To validate the CAM technique in a field situation images of live sponges were captured *in situ*. The test involved photographing four individual sponges with differing morphologies (Fig. 6). The sponges were then collected from the substrate so that an estimate of their true volume could be determined by water displacement. Sponges were wrapped in a thin

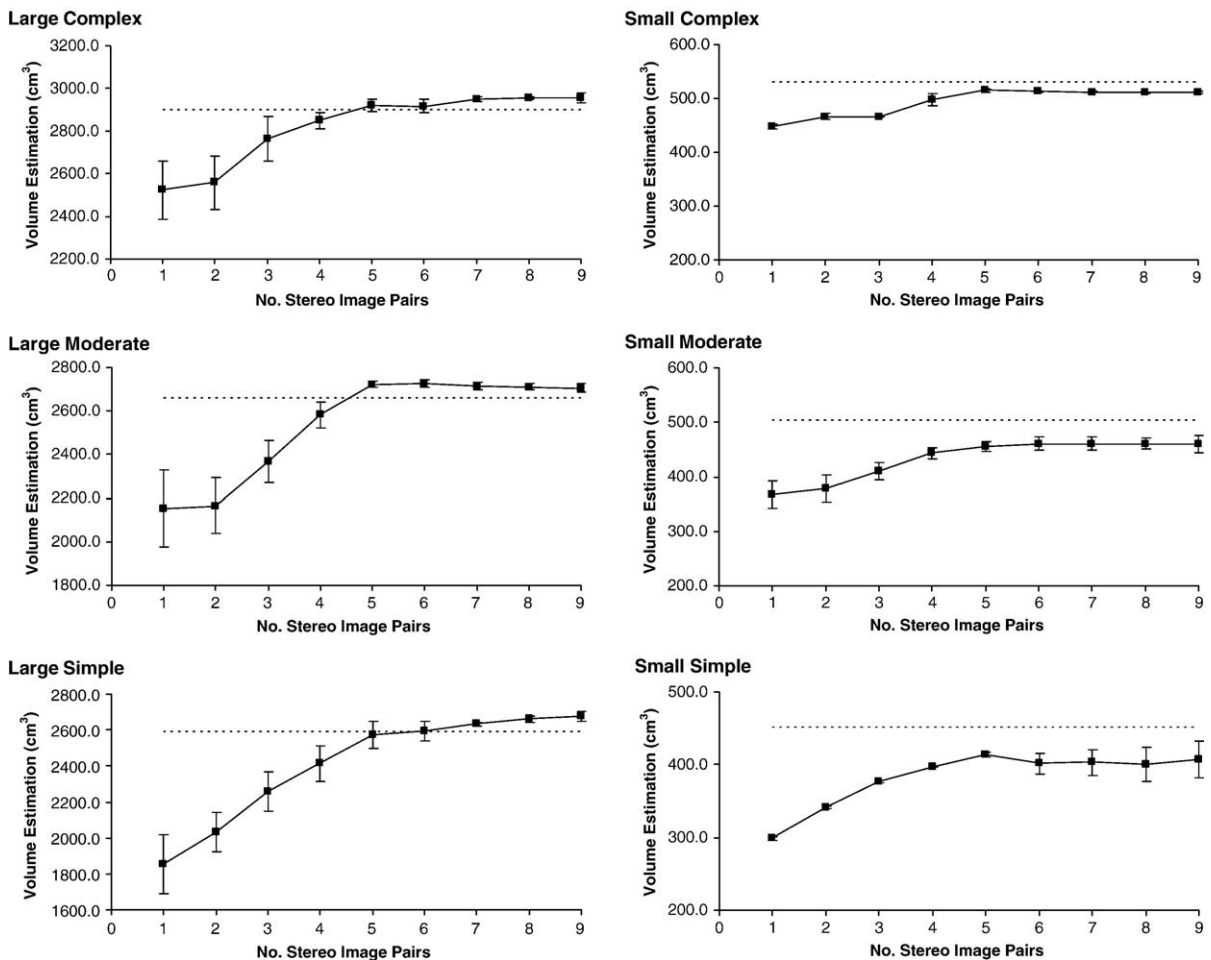


Fig. 7. Mean volume estimates for models showing the asymptotic relationship between the volume estimate and number of stereo image pairs used for each model. Top graphs are complex models, middle graphs show moderate complexity models and bottom graphs show simple models. Note the different scale on the y-axis for each graph (and they do not begin at zero), the dashed line represents the true volume of each model and error bars represent one standard error (see Fig. 5 for models).

Table 1
Results of two sample *t*-test comparing volume measurements made using five and nine stereo image pairs for each model

Model	5 <i>N</i>	9 <i>N</i>	<i>t</i> -Value	<i>df</i>	<i>p</i>
Large complex	9	9	-0.68	All 16	0.51
Large moderate	9	9	0.79		0.44
Large simple	9	9	-1.93		0.072
Small complex	9	9	0.66		0.52
Small moderate	9	9	-0.32		0.76
Small simple	9	9	0.42		0.68

5 *N* and 9 *N* represents the number of replicate volume measurements made using five and nine stereo image pairs respectively.

film of plastic to stop the absorption of water into the sponge tissue and water canals to allow the most accurate measure of their volume by water displacement. CAM \hat{V} were made using the minimum number of stereo image pairs determined to give an adequate \hat{V} , and if the technique proved to be consistent between observers only one observer would be used to make \hat{V} .

3. Results

3.1. Laboratory trials

3.1.1. Number of images

There was a consistent asymptotic relationship between the CAM volume estimate and the number of image pairs used (Fig. 7). For all models, approximately five image pairs were required to

Table 2
One-way ANOVA results for each model comparing observer's volume estimates using five stereo image pairs

Model	Source	<i>df</i>	SS	MS	<i>F</i>	<i>p</i>
Large complex	Observer	2	16740	8370	0.33	0.729
	Error	6	150504	25084		
	Total	8	167244			
Large moderate	Observer	2	4551	2276	0.98	0.427
	Error	6	13896	2316		
	Total	8	18447			
Large simple	Observer	2	98181	49091	4.14	0.074
	Error	6	71069	11845		
	Total	8	169251			
Small complex	Observer	2	225	112	0.56	0.598
	Error	6	1202	200		
	Total	8	1427			
Small moderate	Observer	2	1357	678	3.93	0.081
	Error	6	1035	173		
	Total	8	2392			
Small simple	Observer	2	163	81	0.48	0.638
	Error	6	1008	168		
	Total	8	1171			

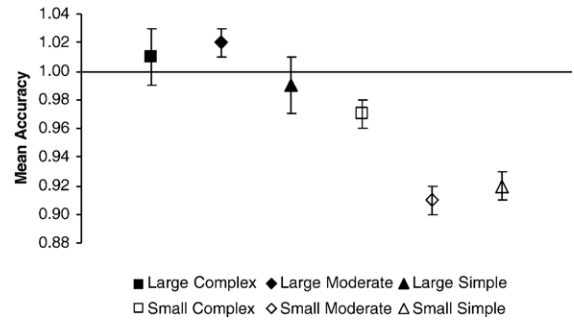


Fig. 8. Mean accuracy of CAM volume estimates for all models. Accuracy is defined as $\mathcal{A} = \hat{V}/V$. Where \mathcal{A} represents estimation accuracy, \hat{V} represents estimated volume and V represents true volume. Accuracy increases as \mathcal{A} approaches one. Volume overestimations are greater than one and underestimations are less than one. Error bars represent one standard error.

produce an adequate volume estimate. This was confirmed by a two sample *t*-test where no significant difference ($p > 0.05$) was seen between the volume estimate made using five and nine stereo image pairs for all models (Table 1).

3.1.2. Technique evaluation

3.1.2.1. Observer bias. Variation between observers' \hat{V} did exist, however, these difference were not statistically significant ($p > 0.05$) (Table 2) for any model. This indicates a high level of agreement between three observers and indicates that the procedure is robust to observer bias.

Table 3
Mean accuracy of the CAM volume estimations (using five stereo image pairs) for each model, and pooled by model complexity (complex, moderate, simple), size (large and small), and the overall accuracy

Model	<i>n</i>	Mean accuracy	Standard error
Large complex	9	1.01	0.02
Large moderate	9	1.02	0.01
Large simple	9	0.99	0.02
Small complex	9	0.97	0.01
Small moderate	9	0.91	0.01
Small simple	9	0.92	0.01
Complex models	18	0.99	0.01
Moderate models	18	0.96	0.02
Simple models	18	0.96	0.01
Large models	27	1.01	0.01
Small models	27	0.93	0.01
All models	54	0.97	0.01

Accuracy is defined as $\mathcal{A} = \hat{V}/V$. Where \mathcal{A} represents estimation accuracy, \hat{V} represents estimated volume and V represents true volume. Accuracy increases as \mathcal{A} approaches one. Volume overestimations are greater than one and underestimations are less than one.

Table 4
Precision of the CAM volume estimations (using five stereo image pairs) for each model, and mean precision

Model	<i>n</i>	Mean \hat{V} (cm ³)	Standard error	Precision
Large complex	9	2920.19	48.19	0.02
Large moderate	9	2722.76	16.00	0.01
Large simple	9	2575.20	48.49	0.02
Small complex	9	515.46	4.45	0.01
Small moderate	9	455.85	5.76	0.01
Small simple	9	413.66	4.03	0.01
Mean				0.01

Precision is defined as $\phi = SE/\bar{x}$. Where ϕ represents volume estimation precision, \bar{x} represents mean estimated volume and SE represents the standard error of volume estimation. Precision increases when SE is small relative to the mean and precision decreases as the ratio increases.

3.1.2.2. Accuracy. Overall, the mean accuracy of CAM's \hat{V} estimates for each model was high with the large complex model being slightly overestimated and large simple model being slightly underestimated ($\mathcal{A}E = 1.01 \pm 0.02$ and 0.99 ± 0.02 respectively) (Fig. 8). The large moderate and small complex models had the next highest level of accuracy, with the large moderate model being overestimated ($\mathcal{A}E = 1.02 \pm 0.01$) and the small complex model being underestimated ($\mathcal{A}E = 0.97 \pm 0.01$) (Fig. 8). The small moderate and small simple models had the lowest accuracy, with both models volumes being underestimated ($\mathcal{A}E = 0.91 \pm 0.01$ and 0.92 ± 0.01 respectively) (Fig. 8). When \hat{V} were pooled by complexity, complex models had the highest accuracy followed by the moderate and simple complexity models (Table 3). Moreover, the CAM technique consistently underestimated the volume of all the small models while large models had a high level of accuracy (Table 3). The pooled

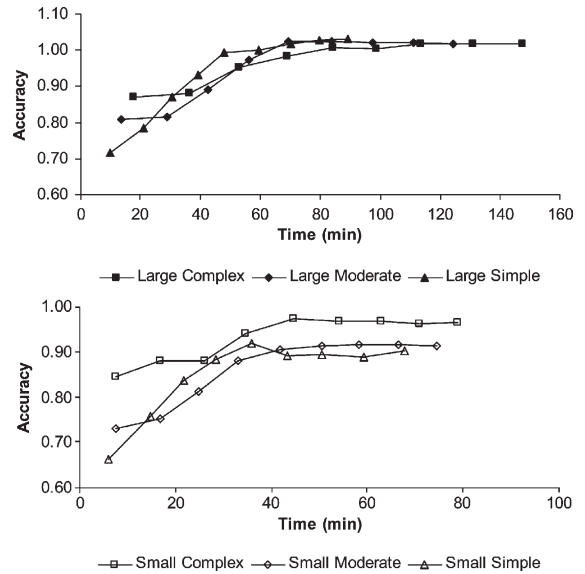


Fig. 9. Graphs depicting accuracy of volume estimation with increasing processing time for an increasing number of stereo image pairs. Top graph shows all large models and bottom graph shows all small models. Accuracy is defined as $\mathcal{A}E = \hat{V}/V$. Where $\mathcal{A}E$ represents estimation accuracy, \hat{V} represents estimated volume and V represents true volume. Accuracy increases as $\mathcal{A}E$ approaches one. Note different y-axes (which also do not begin at zero) and x-axes scales and error bars represent one deviation.

\hat{V} made by the CAM technique for all models had a high accuracy with only a slight underestimation of volume (Table 3).

3.1.2.3. Precision. The precision of the CAM technique was high, with the overall mean precision plus/minus 1% of \hat{V} . The lowest precision was observed for both the large complex and simple models (Table 4),

Table 5
Time taken to complete the necessary operations to make a three-dimensional reconstruction and volume measurement for each model using a minimum number of stereo image pairs (*n*=5)

Operation	Large complex	Large moderate	Large simple	Small complex	Small moderate	Small simple
*Orientation frame positioning			1 min			
*Capturing images			5 min			
Image processing			3 min			
Three-dimensional reconstruction						
Measuring reference points			1 min			
Measuring object points	84 min (±15 min)	70 min (±12 min)	48 min (±11 min)	45 min (±6 min)	42 min (±10 min)	36 min (±11 min)
Volume model generation			2 min			
Total time						
Average	96 min	82 min	60 min	57 min	54 min	48 min
Maximum	111 min	94 min	71 min	63 min	64 min	59 min

Only the time taken to mark points on the object during the three-dimensional reconstruction varies as other aspects are standard procedures. Field based procedures are indicated by an asterisk (*), with all other procedures being performed in the laboratory.

approximately 58.4 cm³ (or 2% of V^*) and 51.5 cm³ (or 2% of \hat{V}) respectively.

3.1.2.4. Efficiency. The large complex model was the slowest to reconstruct with an average time of 1 h 36 min, while the large moderately complex model took an average of 1 h 22 min to calculate \hat{V} (Table 5). The large simple, small complex, small moderately complex and small simple models took a similar amount of time to produce a three-dimensional reconstruction and obtain a volume measurement (1 h, 57 min, 54 min and 48 min respectively) (Table 5). The greatest accuracy and precision for a \hat{V} were achieved with the above time investments for each model (Figs. 9 and 10).

3.2. Field application

The four sponges used to test CAM in a field situation had different morphologies, and had similar sizes and complexity of shape as tested with the plasticine models. The tubular sponge had the lowest level of surface complexity and approximates the moderately complex and simple plasticine models. The flabellate sponge and both massive sponges approximate the moderately complex and complex models, with multifaceted surface features. Lowest \hat{V} accuracy was observed with the

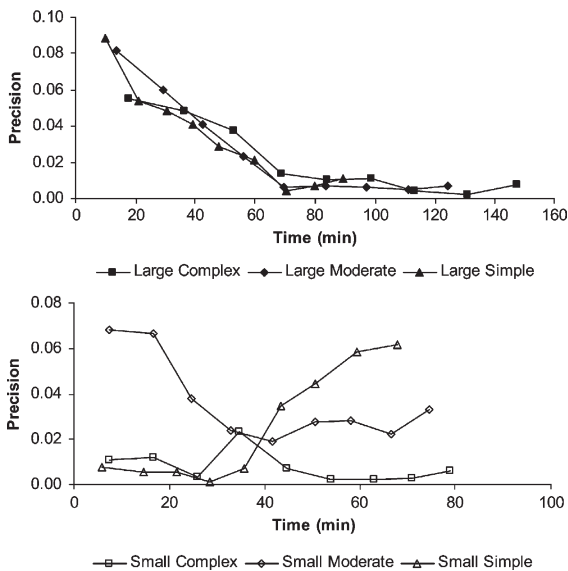


Fig. 10. Graphs depicting increasing precision of volume estimation with increasing processing time for an increasing number of stereo image pairs. Top graph shows all large models and bottom graph shows all small models. Precision is defined as $\phi = SE/\bar{x}$. Where ϕ represents volume estimation precision, \bar{x} represents mean estimated volume and SE represents the standard error. Precision increases as the ratio decreases. Note different x-axis scales.

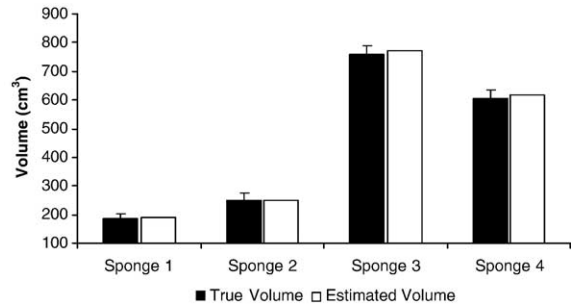


Fig. 11. Graph depicting the accuracy of CAM’s volume estimate to the true volume of four real sponges to test the technique’s field application. Note y-axis does not start at zero and error bars represent one standard deviation.

massive sponges 1 and 2, with the \hat{V} of each sponge being overestimated ($\mathcal{E}=1.02$ and 1.02 respectively) (Fig. 11). The highest accuracy in \hat{V} was with the tubular sponge, with its volume being slightly underestimated ($\mathcal{E}=0.99$) (Fig. 11). The flabellate sponge’s \hat{V} was also accurate and slightly overestimated ($\mathcal{E}=1.01$) (Fig. 11). The mean accuracy of \hat{V} for all four sponges was high ($\mathcal{E}=1.01$), as was the mean precision for \hat{V} ($\phi=0.01$).

The time taken to make a three-dimensional reconstruction was also recorded for each sponge, with the first massive sponge taking longest (65 min) to obtain a volume measurement. The tubular sponge was the quickest to process (35 min), while the second massive sponge and flabellate sponge took a similar amount of time (53 min and 46 min respectively). These times are comparable if not quicker than the times taken during three-dimensional reconstruction of the plasticine models.

4. Discussion

The stereo-photogrammetric technique we have developed and trialled allows for low observer bias; it is accurate, precise and provides efficient volume measurements of morphometrically complex sessile epibenthic invertebrates. The method was primarily developed to obtain measurements of volume for quantitative studies of growth in sponges, however, associated parameters such as surface area and morphometric details (such as length, width and height) are also easily determined with this technique. The method represents a significant advancement in the use of photogrammetry on benthic organisms underwater due to its simplicity, ease of use, low expense, and efficiency (both during processing and underwater during image capture) which is critical for marine based research.

The volume estimates made using the CAM technique did not reveal any significant difference ($p>0.05$)

between observers suggesting this technique provides repeatable measurements not biased by the software user. However, a bias within the technique was observed when producing a volume estimate for small objects, which were consistently underestimated for the small models in the laboratory trial. The inaccuracy seen with these models can be related to the sharpness (tight curvature) of the models' edges as a result of the software having insufficient overlap between the triangulated surfaces from the various image pairs. This can be resolved by adjusting the separation of the two cameras of the stereo system to suit the size of the object that is being measured. For example, when measuring smaller objects a smaller camera separation would increase the measurement accuracy and ability, whereas, with larger objects a greater base separation would be beneficial. These modifications relate both to the level of image overlap and the camera base selection which are a trade-off between the size of the object and range to the targeted object(s) (Eleftheriou and McIntyre, 2005).

Accuracy was greater on complex objects that approximate the morphological complexity to be expected in nature. On simpler subjects accuracy decreased and this result can be related to both the lack of surface features on the simple and moderately complex models. This lack of features limits the creation of a sufficient point cloud for accurate three-dimensional reconstruction by the software, resulting in an underestimation of the simple and moderate models. However, it is highly unlikely that a biological organism will have a morphology as simple as the simple and moderate models tested in this study.

The accuracy of CAM volume estimations was comparable and higher than that reported by the only previous (Cocito et al., 2003) study using photogrammetry to determine volume of benthic organisms. However, a true comparison cannot be made as similarities between the complexities of models are not known. Cocito et al. (2003) also reported a similar pattern of accuracy for the different levels of complexity reported in this study, that is, complex models were overestimated and simple models underestimated.

CAM volume estimates were also found to be highly precise in this study (lowest precision $\pm 2\%$) and this figure is comparable to the precision reported by Cocito et al. (2003).

The efficiency of volume estimates represents the most significant benefit of the CAM technique with the longest mean processing time for a volume estimate (from the large complex model) approximately 50% faster than the time reported by the only comparable technique, MOD3D (Cocito et al., 2003). Most of the

time taken to produce a volume estimate in CAM is consumed by point measurement, with the actual volume generation taking less than 1 min. This is encouraging as there are numerous established photogrammetric techniques that can be used to increase the efficiency of point measurement in CAM. This is an area of ongoing software development.

In terms of a field technique, the CAM method was very accurate with the lowest accuracy recorded for the two massive sponges (massive 1 and 2), which is a small overestimation. The slight inaccuracy recorded for the field could be attributed to epiphytic organisms growing on the target sponge which was particularly the case with the first massive sponge (massive 1), thus making it difficult to accurately mark surface features. Inaccuracy was further increased by the inability to delineate the substrate/organism boundary due to obstruction by surrounding organisms. This is exemplified by the flabellate and tubular sponges where accuracy increased because of the limited obstruction from surrounding organisms as well as the lack of epiphytic organisms on their surface. The only comparable study (Cocito et al., 2003) did not sample *in situ* specimens during the testing of their technique, thus a comparison in terms of field applicability cannot be made.

CAM is highly accurate (even when taking into account the largest mean absolute inaccuracy ($\pm 9\%$) seen in this study for the small moderate model) when compared to the errors reported for other techniques used to study sponges. For example, Leys and Lauzon (1998) reported an error of 13% when studying the growth of Hexactinellid sponges using a single camera photographic method, and Duckworth and Battershill (2001) reported an inaccuracy of approximately 15% using a similar single camera method.

The method presented here is non-invasive, does not require any reference markers or tags fixed to the object being modelled (for referencing and/or calibration) which is likely to cause damage to the target organism hindering any monitoring studies. Although this study employed the use of a reference frame during the capturing of images, it is not used for calibration, but as an aid to speedup the three-dimensional reconstruction process. A reference frame does allow for any directionality of volume change to be determined if patterns of growth were being investigated, as subsequent volume determinations could be made in the same orientation as the initial measurement.

The advancements made with the CAM technique have resulted in an uncomplicated, quick and easy to use underwater measuring method for complex benthic organisms. This method can be utilised in a greater

variety of studies, particularly in research outside the limits of SCUBA (Hancock and Milner, 1986), as the system can be mounted on an ROV or submersible without the need for any additional equipment. CAM also allows for precise and accurate measurements of volume and surface area for ecophysiological studies (for example energy flow) (Edgar, 1990; Eleftheriou and McIntyre, 2005), the monitoring of growth or partial mortality, as well as other ecological processes (e.g. overgrowth/competition), and has applications in aquaculture for monitoring culture success.

The CAM technique is not without limitations, but similar limitations hinder any photogrammetric technique for studying benthic organisms. CAM volume estimations of benthic organisms will always have an inherent error associated with the inability of photogrammetry to discern the volume of internal spaces in an organism. This is particularly the case in sponges, however, if the internal volume is known (examined by determining average internal space of the target organism) this could be factored into any volume estimations. Additionally, like all photogrammetric techniques dealing with benthic organisms, it is not always possible to discern the structure of the substrate below the target organism. This is only an issue for initial measurements, as the substrate is unlikely to change and subsequent measurements would reveal any changes. Turbidity of the water column also hinders any underwater technique employing photogrammetry, but this is less of an issue when studying individual organisms as the subject to camera distance is relatively small, lessening the amount of suspended material between the subject and camera.

Future advancements and refinements of this technique will increase its accuracy, precision, and efficiency. Software developments could further automate the three-dimensional reconstruction of an organism, the inclusion of a seed point laser array (produces a grid of laser dots on the subject) will further quicken and increase the accuracy of this technique. Advancements in camera technology will benefit the use of photogrammetry as a sampling tool by providing high quality, high resolution images, and better optical properties.

The technique described in this study has clearly demonstrated the unbiased, accurate, precise and efficient measurement of morphologically complex benthic organisms using photogrammetry. The technique is robust to variations in user experience, removing any observer biases and results in accurate and precise measurements. Underwater photogrammetry and associated three-dimensional reconstruction has many advantages over traditional sampling techniques, and it is clear that this technique will be beneficial in the research of benthic organisms.

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