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Authors: Błażejowski, Błażej, Binkowski, Marcin, Aleksandra Bitner, Maria, and Gieszcz, Piotr

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Technical note

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X-ray microtomography (XMT) of fossil brachiopod shell interiors for taxonomy

BŁAŻEJ BŁAŻEJOWSKI, MARCIN BINKOWSKI, MARIA ALEKSANDRA BITNER, and PIOTR GIESZCZ

The ability to see and understand the three-dimensional structure of an investigated object plays a key role in studying fossil remains. All living organisms are formed in three-dimensions, but unfortunately fossilization processes often reduce overall shape, making it difficult to gather information about real overall appearance, functionality, and inner structure. Here, using a specimen of the brachiopod *Terebratula terebratula* we demonstrate a non-destructive technique for exploring the 3-D internal structure of fossil remains. The use of tomography allows the construction of a set of transverse serial sections in the manner used by brachiopod researchers for decades.

Introduction

The common method for studying the inside of 3-D fossil brachiopods has been serial sectioning (Wiśniewska 1932), which is not only a time-consuming technique, but also irreversibly destroys investigated specimens. Despite many disadvantages, destructive methods like serial sectioning or serial grinding (e.g., Sutton et al. 2005) are considered by brachiopod researchers as the only possibilities to reveal very important details of inner morphology of investigated fossil taxa.

The main goal of this report is to describe the application of X-ray microcomputed tomography (XMT) as a noninvasive tool for detailed research on fossil marine invertebrate faunas. The method provides excellent results in high-resolution quantitative volumetric investigation on diverse samples.

In our studies we used a single specimen of the short-looped terebratulide brachiopod (Fig. 1A) *Terebratula terebratula* (Linnaeus, 1758) from the Pliocene of Velerín, southern Spain (see Bitner and Martinell 2001). To present a 3-D model with virtually cut out part of the ventral valve (Fig. 1B) the XMT technique was applied, which is based on differences in attenuation of the X-ray beam propagating through a solid object. Background was removed from the images by thresholding, and a number of virtual transverse cross-sections was generated (Fig. 2) to show internal structure of the specimen. Further cross-sections were generated based on resliced original data. The result is similar to traditional sectioning obtained by destructive slicing with the resolution defined by voxel size reaching here 22.4 × 22.4 × 22.4 μm.

The advantages of XMT being a non-destructive technique and the resultant data on internal structures together with novel computer techniques like segmentation or outer layers transparency, should be considered as essential for identification and comparisons within assemblages of poorly preserved or damaged specimens.

Discussion

The most significant disadvantage of traditional, invasive methods such as serial sectioning is very limited information about volumetric dependencies within the internal structure. Some part of the material is irreversibly lost during preparation, while X-ray tomography allows the generation of serial sections in any cutting plane, a set of planes spaced almost every slice thickness (in our case equal to 22.4 μm). It provides the opportunity for detailed analysis of the brachidium and cardinalia. The computed image is so clear and of such good quality, that it is even possible to study microfossils preserved in matrix inside the brachiopod shell (see SOM, the Supplementary Online Material at http://app.pan.pl/SOM/app56-Blazejowski_etal_SOM.pdf). One of the most effective ways of digital processing and analyzing tomographic data is the construction of isosurface-based "virtual fossils", that can be manipulated and dissected interactively (Sutton 2008).

Recently studies on brachiopods using SkyScan 1172 X-ray microtomography have been made (Pakhnevich 2010), but to

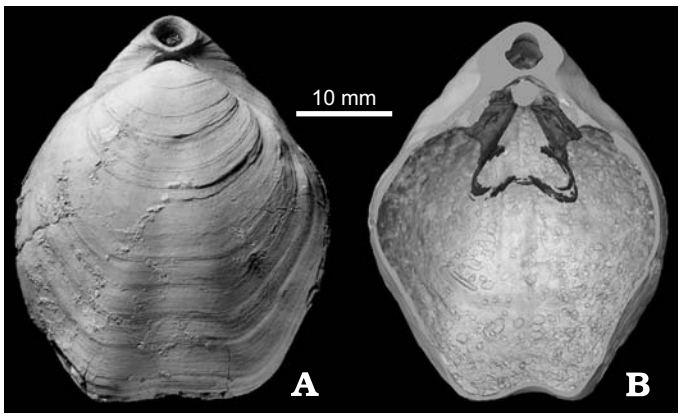

Fig. 1. Terebratulide brachiopod *Terebratula terebratula* (Linnaeus, 1758), Pliocene, Velerín, Spain; complete specimen, ZPAL Bp.XLIV/9. **A**. Dorsal view. **B**. Reconstruction of 3-D internal structure with virtually cut out part of the ventral valve.

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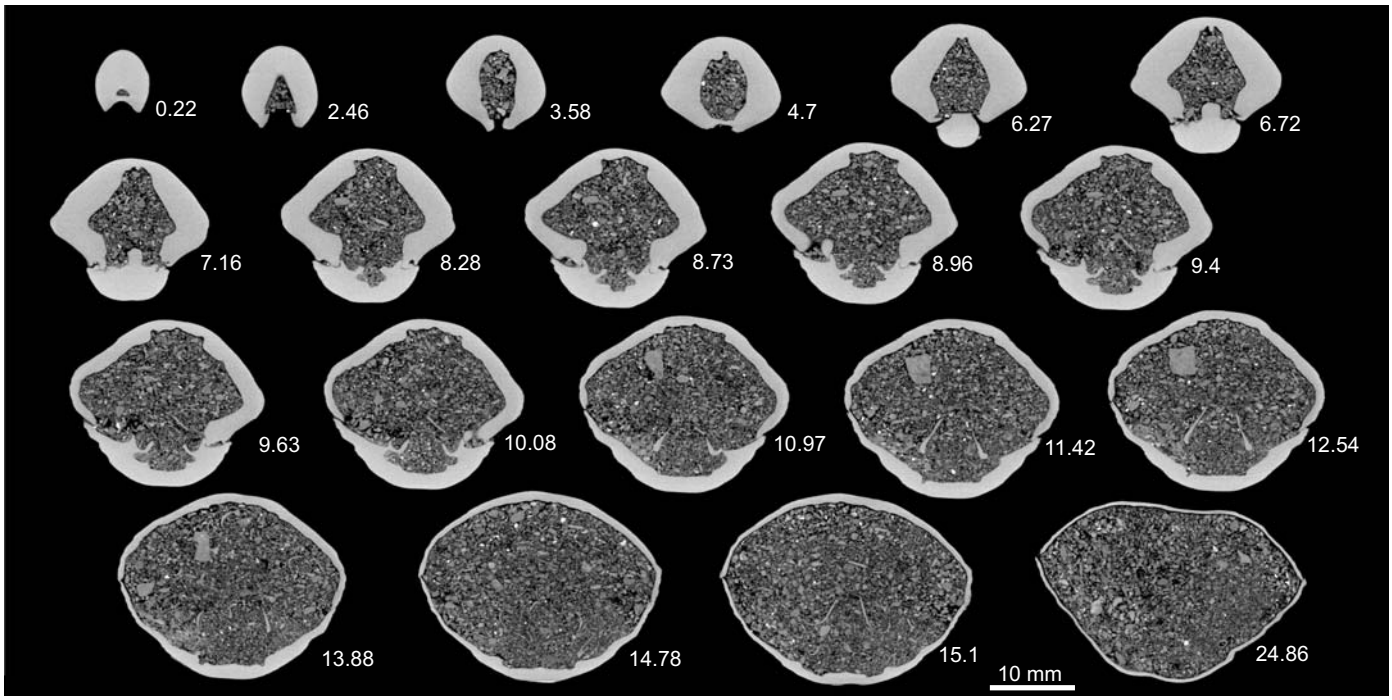

Fig. 2. Transverse serial sections of *Terebratula terebratula* (Linnaeus, 1758), through specimen ZPAL Bp.XLIV/9; Pliocene, Velerín, Spain. Numbers indicate distance in mm from the tip of the ventral umbo.

date virtual cross-sectioning and detailed 3-D reconstruction of the complete brachidium was unsuccessful in fossil specimens.

In this report we present the brachidium and hinge structures revealed using Benchtop (Nikon Metrology, Tring, UK) XMT scanner on a fossil specimen. Visualization (3-D model) of the brachidium is shown in SOM (http://app.pan.pl/SOM/app56-Blazejowski_etal_SOM.pdf). These internal structures are critical for classification, especially in this group of animals, as well as for understanding their phylogeny, thus the method should be very helpful in cases where traditional sectioning cannot be used.

Choosing the worst specimens for destructive investigation is a common practice, as the best preserved ones are held for holotypes (Alvarez and Brunton 2008). The use of this new and advanced method enables the use of holotypes in order to check their conspecificity with the sectioned paratypes (Motchurova-Dekova and Harper 2010). In such case, choosing the best specimens (holotypes) for non-destructive XMT studies should become common practice. Furthermore, XMT constructed virtual specimens can be a subject of various studies including collaboration with other scientists. Virtual fossils can be accessible on-line, enabling cooperation among geographically isolated researchers.

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Błażej Błażejowski [bblazej@twarda.pan.pl] and Maria Aleksandra Bitner [bitner@twarda.pan.pl], Institute of Paleobiology, Polish Academy of Sciences, ul. Twarda 51/55, PL-00-818 Warszawa, Poland;
Marcin Binkowski [marcin.binkowski@us.edu.pl], Department of Biomedical Computer Systems, University of Silesia, ul. Będzińska 39, PL-41-200 Sosnowiec, Poland;
Piotr Gieszcz [p.gieszcz@twarda.pan.pl], Association of Polish Climatologists, ul. Międzynarodowa 58/36, PL-03-922 Warszawa, Poland.

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