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Interactive comment on "Non-invasive imaging methods applied to neo- and paleontological cephalopod research" *by* R. Hoffmann et al.

Anonymous Referee #1

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Summary of the paper: As the title of their paper says, Hoffmann et al. studied and wrote about, "Non-invasive imaging methods applied to neo- and paleontological cephalopod research." The application of non-invasive methods in paleontology more generally has been used and is of great interest, given the nature of and possible rarity, value and condition of fossil specimens. Hoffmann et al. are interested in such methods being applied to cephalopod studies, and in particular, to ammonoid research.

The paper describes different imaging methods: surface scanning instrumentation, computed tomography (CT) techniques at different scales and resolutions (medical, micro-, nano-), magnetic resonance imaging (MRI), and synchrotron radiation micro-computed tomography (SRμCT) with regard to sample size needed, accessibility to and ease of instrumentation usage, environment to house instrumentation, data acquisition

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(quality and amount) and storage availability, scan time duration and data analysis capabilities, compatibility with other platforms and software for post-processing and visualization, and cost of scans and instrumentation . In particular, the authors illustrate applications to ammonoid research via shell volume that can be used in a buoyancy calculation as a way to measure swimming capacity, ontogeny as inferred by the shell's chamber volume, and revealing obscured morphological characters. The potential for making formerly obscure morphological characters visible is useful in applications that utilize descriptive morphology and morphometry.

The authors have shown as others have that 3D imaging and reconstruction can be transformative in a discipline such as paleontology, where analysis and our understanding about life on Earth is reliant on collecting information and data on organismal morphology. In their paper, the authors recognize this and have added to the body of knowledge that will aid in paleontology becoming more comprehensive, quantitative, and analytically sophisticated. However, there are some problems with the paper to be noted and discussed, and that the matters raised need to be addressed by the authors.

Questions and comments about the paper: The abstract is incomplete and not entirely representative of the paper. In the abstract, the authors state that the main application of imaging methods they describe is used in "morphometry and volumetry of cephalopod shells in order to improve our understanding of diversity and disparity, functional morphology and biology of extinct and extant cephalopods." Diversity and disparity are not addressed or discussed at all in the paper, and functional morphology is only inferred by the measurement of shell volume. In the text, no attempt is made to describe or discuss how volume is used in a biomechanical context, except the authors do cite Anderson et al 2012 with obliquely mentioning, but not explaining, how to use imaging techniques with regard to, for example, finite element analysis. The authors describe and calculate volume and use this to represent buoyancy. How is buoyancy related to a dynamic behavior such as swimming (including propulsion) vs. a static behavior such as floating? Further, there is no formula given on how volume or buoyancy was calcu-

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lated and no references are cited to this effect, most notably, there was no mention of Raup and Chamberlain (1967). It is a bit of a stretch to say that functional morphology was covered in the text. In addition, although it was mentioned in the text a number of times, the morphological species concept was not mentioned in the abstract.

Concerning the morphological species concept, how does using imaging techniques "sharpen," "contribute to," "improve " (the authors' words) the "morpho-species" concept? The imaging techniques have nothing to do with the concept; rather, the methods are another way to describe morphological attributes or characters or enable calculations for morphometry.

The authors state throughout their paper that they are making comparisons among different imaging techniques. They do compare the same categories/specifications of information for all the instrumentation being reviewed (see above summary of the paper). However, when it comes to actual application to research, deciding which imaging method to use depends on the question of interest by the researcher, the scale of the morphology of interest, and the particular resolution and specifications of the instrument rather than making a comparison of multiple kinds of instrumentation. For example, the authors obtained the best images for the morphology of septa and spacing with micro-CT (resolution of 7.5μm), moment of hatching with nano-CT (resolution of 1.0 μm), and secondary calcite crystals with SRμCT (resolution of 0.74 μm). This illustrates that three different research questions are at work here, and three different instruments were appropriately used. One could argue that an actual comparison among methods when specifically applied to research is non-existant. I think a more accurate assessment of the contributions of this paper would be that the authors compared various imaging instrumentation specifications, showed how non-invasive methods could be used to acquire data on morphological features, and that volume can be calculated with the aid of such non-invasive techniques. I think the comparisons in terms of research are incidental and reflect more of an artificial framework in which to present results.

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I would have like to have seen more quantitative information on 3D reconstruction and associated problems and error measurements for each of the imaging methods. In addition, I would have liked more information on other software packages and post-processing as well as data storage, access and sharing with regard to high performance and cloud computing. For example, some of the issues the authors identify with some of the instrumentation would disappear with consideration of the use of high performance computing.

Some of the paper is devoted to ontogeny, including Figures 6 and 7. In Fig. 6, the numbering of chambers in illustration 6A seems to be the opposite of what is used in Fig. 6D, which is a plot apparently using the numbering system from Fig. 6A. In Fig. 6A, the smaller the chamber size, the higher the chamber number, and therefore this is nearer to the protoconch and is representative of older growth. In Fig. 6D, the higher the chamber number, the higher the volume which is the most recent growth of the ammonoid. In Fig. 6D, the x-axis needs to start at chamber 35 and decrease to chamber 1, or perhaps, is the label for the y-axis supposed to be cumulative volume? In any event, the graph in Fig. 6D is confusing. Although Fig. 6A is the same as 6B, it has been changed with the colors, markings and numbers layered over it. That is fine; however, Figs. 6B and 6C are the same as a previous publication. This is not noted in the text or figure legend.

Why are the plots in Fig. 7 semi-log plots? Since the maximum diameter is 17 cm, is it necessary to have a scale on the x-axis to 100 cm? Both in the text and in the Fig. 7 legend, there is no discussion of isometry and allometry with respect to growth and ontogeny. The authors' discussion is limited to quantification of changes in ontogeny as chamber volume changes. How should these changes be quantified and compared between taxa? Between extant and extinct ammonoids? What assumptions and models of growth can be used to formulate changes in ontogeny with respect to isometry and allometry?

Although preservation was obliquely mentioned, there was no direct discussion about

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how to use imaging techniques to quantify or characterize taphonomy. That is, since more detail can be obtained with imaging techniques, how are these techniques applicable in taphonomic analysis? Using imaging techniques, how does taphonomy quantitatively affect morphological assessment?

The authors tell us how non-invasive methods potentially eliminate damage to specimens. This is true. How are non-invasive methods useful with respect to minimization of contamination of specimens? Is this so, or if not, why not?

In general, the authors present how imaging methods can be used in morphological analysis. Aside from the non-invasive nature of the methods they use, how would analyses using imaging and 3D reconstructions be done that would match or exceed the detailed results of work done by, for example, Klug (2001) or Kröger (2002), or for that matter, by those who do detailed traditional work? If the authors could take the time to make this comparison, it would show cephalopod researchers not only the utility of imaging methods, but also potentially the superiority of imaging methods. By including analyses and text addressing this issue, the paper would be greatly improved. For example, the authors' illustration of ontogenetic changes in attachment scars using nano-CT and SRμCT in Fig. 9 is a good start, but more needs to be said here.

Some references (not a complete list) that should be cited and their contents incorporated into the text: Klug, C. 2001. Life-cycles of some Devonian ammonoids. Lethaia 34: 215-233.

Klug, C. and Korn, D. 2004. The origin of ammonoid locomotion. Acta Paleontologica Polonica 49: 235-242.

Kröger, B. 2002. On the efficiency of the buoyancy apparatus in ammonoids: evidences from sulethal shell injuries. Lethaia 35: 61-70.

Lukeneder, A. 2012. Computed 3D visualisation of an extinct cephalopod using computer tomographs. Computers & Geosciences 45: 68-74.

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Raup, D. M. and Chamberlain, Jr., J.A. 1967. Equations for volume and center of gravity in ammonoid shells. Journal of Paleontology 41: 566-574.

* Work by the following authors could be cited as well: W.B. Saunders; E.J. Denton; J.A. Jacobs.

Technical aspects of the paper: Some of the problems encountered: Throughout the paper, commas are absent from places where conjunctive and adverbial clauses should be separated. There are places in the paper where the wrong term or awkward sentence structure is used, and this makes interpretation of the text difficult or confusing. There are places where plurals should be used instead of the singular. Throughout the text "weight" is used when the correct term should be "mass." There are typos and lack of appropriate punctuation throughout the paper. Overall, a thorough job of careful editing is required to bring the text (including the figure legends) up to an acceptable standard.

There are inconsistencies and omissions as well. Figs. 7 and 8b are not cited in the text. Figures cited in the text that are not in the proper order: Fig. 3b is cited before Fig. 2; Fig. 5a is cited before Fig 4. In the "Material" (should be "Materials") section, all taxa used in the authors' studies are listed by name. However, in the "Introduction" section, Fig. 1 is mentioned in the text, but the name of the taxon, Gaudryceras, is not. For Hibolithes, no mention is made in the text (section 4.6), although the taxon is mentioned in the figure legend for Fig. 11. In the legends for Figs. 6 and 7, no mention is made that the images were taken with micro-CT. For Fig. 12, the legend refers to "growth" and "shrink" models, but no explanation is given in the text.

Interactive comment on Biogeosciences Discuss., 10, 18803, 2013.