Supplementary materials for

## 3D revisualisation: a new method to revisit segmented data

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# Supplementary Notes

### Drishti v2.7

### Installation

Drishti v2.7 is available to download from https://github.com/nci/Drishti under "Releases".

When you click, it will lead to a version-specific webpage where the zip file is located at the bottom of the page; click *Drishti* v2.7.zip to download *Drishti*.

Source code and detailed information about this new release are also available on GitHub. Please note that *Drishti* v2.7 can only run on the Windows operating system. However, users can compile and install *Drishti* v2.7 for CentOS/Ubuntu. Sample compilation script for \*nix systems is provided to users with the source code.

Once the download is finished, unzip the .zip file. Go to the "bins" folder; then, you should be able to run *Drishti* v2.7. *Drishti* is designed as a portable application, thus making sure you know where the portable directory is stored on your computer.

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# Supplementary Figures



Supplementary Figure 1. Ventral view of the segmented braincase of fossil fish *Tungsenia* IVPP V10687. a-c, Extracted images from Mimics. a, Original image output from Mimics. b, 8-bits greyscale image of a. c, Detected edges of b. d, 2D histogram of b. e, the 3D surface plot of b. f-h, Extracted images from *Drishti* after revisualization. f, Original image output from *Drishti* in the same orientation and scale of a. g, Revisualized result corresponds to b. h, Revisualized result corresponds to c. i, 2D histogram of g. j, the 3D surface plot of g.



Supplementary Figure 2. Lateral view of segmented fossil bird *Linxiavis* IVPP V24116. **a-c**, Extracted images from Avizo. **a**, Original image output from Mimics. **b**, 8-bits greyscale image of **a**. **c**, Detected edges of **b**. **d**, 2D histogram of **b**. **e**, the 3D surface plot of **b**. **f-h**, Extracted images from *Drishti* after revisualization. **f**, Original image output from *Drishti* in the same orientation and scale of **a**. **g**, Revisualized result corresponds to **b**. **h**, Revisualized result corresponds to **c**. **i**, 2D histogram of **g**. **j**, the 3D surface plot of **g**.



Supplementary Figure 3. Ventral view of the segmented upper jaw and inner ear of lizard Varanus indicus (AMNH R58389). a-c, Extracted images from Mimics. a, Original image output from Mimics. b, 8-bits greyscale image of a. c, Detected edges of b. d, 2D histogram of b. e, the 3D surface plot of b. f-h, Extracted images from *Drishti* after revisualization. f, Original image output from *Drishti* in the same orientation and scale of a. g, Revisualized result corresponds to b. h, Revisualized result corresponds to c. i, 2D histogram of g. j, the 3D surface plot of g.



Supplementary Figure 4. Images used for optical density measurements. All images are from segmented volumes using Mimics and their corresponded revisualized segmented volume data using Drishti. a-d, Extracted images of *Tungsenia* IVPP V10687. a-b, Ventral view, from segmented volume (a) and revisualized segmented volume (b). c-d, Dorsal view, from segmented volume (c) and revisualized segmented volume (d). e-h, Extracted images of *Erofoichthys* IVPP OV2715. e-f, Ventral view, from segmented volume (e) and revisualized segmented volume (g) and revisualized segmented volume (h).



Supplementary Figure 5. The median optical density of images from Supplementary Fig.4. Calibration of optical density was completed using an 8-bits 21 steps-table using ImageJ. Data analysis was carried out in Microsoft Excel.



**Supplementary Figure 6. Correlation between the median optical density of images in Supplementary Fig.4.** The X-axis represents the median optical density of images obtained from the original segmented volume using Mimics (i.e. Supplementary Fig.4a,c,e,g). Y-axis represents the median optical density of images obtained from the revisualized volume using *Drishti* v2.7 (i.e. Supplementary Fig. 4b,d,f,h). Analysis was carried out in Microsoft Excel.



**Supplementary Figure 7. Step table and calibration curve were used for this study.** a, step-table. b, calibrated Optical Density curve with the correlation coefficient R<sup>2</sup> equals 0.9990.