VisuAlign Nonlinear tool

Contents

1. Introduction

1.1 Background

VisuAlign is a tool for applying user-guided nonlinear refinements (inplane) to an existing, affine 2D-to-3D registration, such as created using QuickNII (https://www.nitrc.org/projects/quicknii/).

While linear registration tools are vital in bringing experimental image data to standardized coordinate spaces, for precise quantitative analysis one has to address the residual anatomical variability among test subjects after registration. VisuAlign is a tool designed for this task.

Rodent atlas delineations provided with VisuAlign:

- Waxholm Space Atlas of the Sprague Dawley rat brain, versions 2 and 3 (at 39 µm resolution)

- Allen Mouse Brain Atlas reference atlas, CCFv3 (delineations from 2015 and 2017 at 25 µm resolution).

> **For example of use, tutorials and demo video, Please visit :**
> **https://ebrains.eu/service/quicknii-and-visualign**

1.2 Condition of use

VisuAlign v0.8 is developed by the Neural Systems Laboratory, Institute of Basic Medical Sciences, University of Oslo, Norway, with support from the European Union Seventh Framework Programme (FP7/2007-2013) under grant agreement No. 650003 (HBP FPA).

Use of the VisuAlign software is allowed provided that reference is given to the following:

- resource identifier: VisuAlign, RRID:SCR_017978
- publication: Puchades MA, Csucs G, Lederberger D, Leergaard TB and Bjaalie JG. Spatial registration of serial microscopic brain images to three-dimensional reference atlases with the QuickNII tool. PLosONE, 2019, 14(5): e0216796.

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1.3 Contact

For technical inquiries and user support, please visit https://ebrains.eu/service/quicknii-and-visualign.

1.4 Requirements

VisuAlign uses files generated for and by QuickNII. VisuAlign therefore has the same image requirements as QuickNII:

Standard web-compatible image formats are supported, 24-bit PNG and JPEG. Images can be loaded up to the resolution of 16 megapixels (e.g. 4000x4000 or 5000x3000 pixels). However, just like QuickNII, VisuAlign does not benefit from image resolutions exceeding the resolution of the monitor in use. For a standard FullHD or WUXGA display (1920x1080 or 1920x1200 pixels) the useful image area is approximately 1500x1000 pixels, using a similar resolution ensures optimal image-loading performance and also eliminates excess storage size. As fulfilling these requirements usually requires preprocessing of the images (converting to PNG or JPEG and downscaling to screen-like size), VisuAlign keeps track of original image dimensions as part of its series descriptor.

2. Open VisuAlign and load the data

- Use the same folder as QuickNII with your png images and series descriptor (xml file). You will need the JSON version of the descriptor (it is generated in QuickNII by clicking **"Save JSON"**).
- Open tool by clicking on "VisuAlign.bat" file. It loads and saves JSON files created by QuickNII, →File, Open, and choose your JSON file.

  Note: The JSON file generated by VisuAlign can be opened in QuickNII: the additional nonlinear marker points will not be deleted. Save new adjustments in JSON format only.
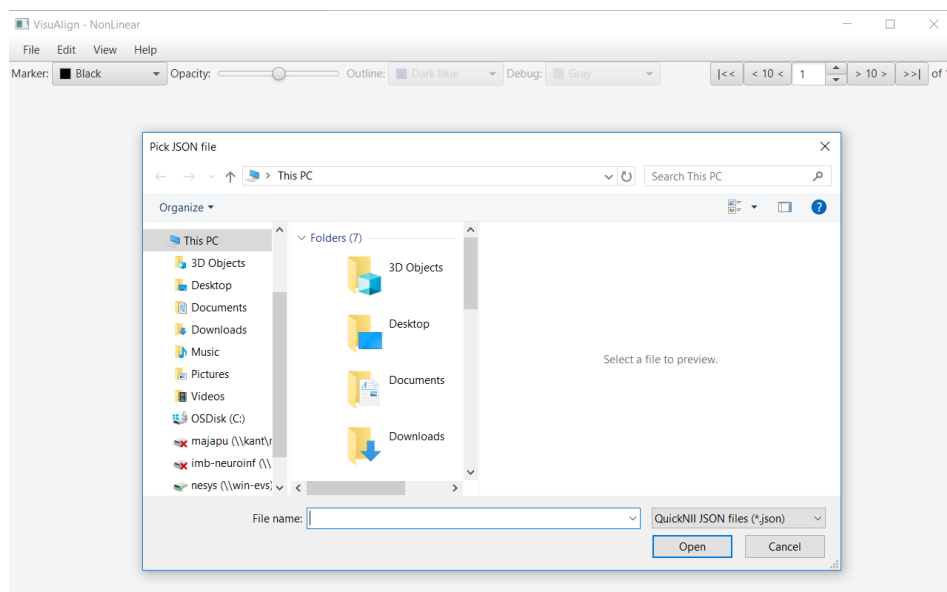
3. Navigating between sections

- Navigating between slices happens in the top-right corner, jump to first section (|<<), jump 10 sections towards the start (<10<), and the same towards the end. Keyboard shortcuts for navigation are the Left/Right arrows, moving one section back/forth.
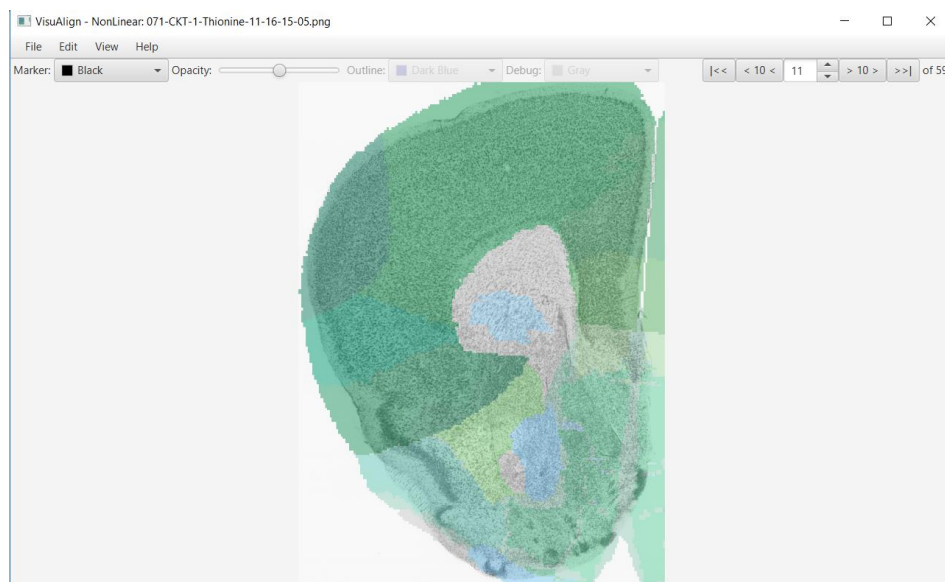
4. Working with markers

- New markers are added under the mouse cursor by pressing the Space bar
- Nonlinear distortions are applied by dragging a marker using the mouse
- Press Delete or Backspace to remove a marker under the mouse cursor

- A good strategy is to place markers around the contour of the section first, and then proceed with refining the inner parts.
- Markers can and should be used to pin/surround locations that should not be affected by further modifications.
- The default marker color is black, but it can be changed using the first color picker (near to the top-left corner of the application window)
- Try to keep the number of markers to a minimum.
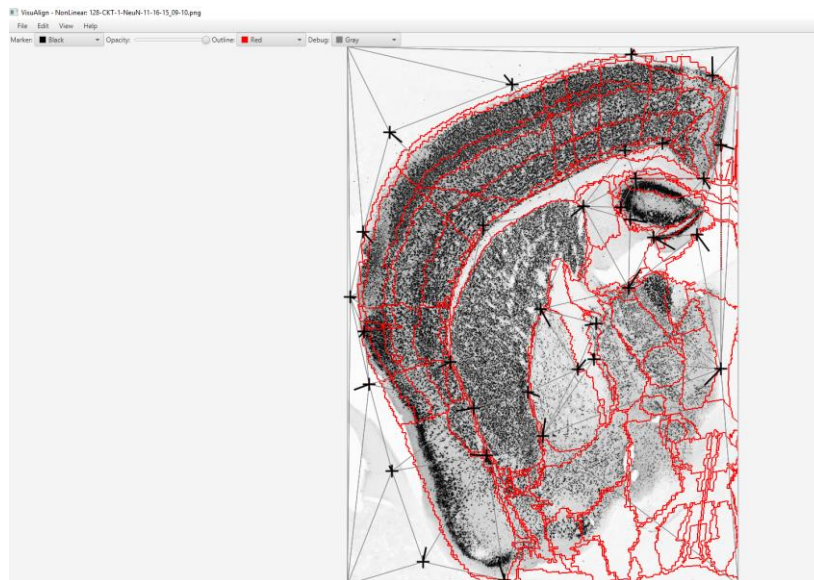
- The outline mode is toggled by pulling the opacity slider to the far right (as seen in the screenshot). A color picker becomes active for changing the outline color.
- "Debug mode" displays triangles representing the deformation field: it is enabled from the View menu (and also enables a color picker for changing the color of the triangles)

5. Saving results and export

- "Save As" saves the deformations in a new JSON file. This file is compatible with Nutil Quantifier for QUINT analysis.
- There is also an "Export" function for generating .flat files for Nutil (.png-s are created too, and right now it exports only the sections, which actually have non-linear refinements).
- The export also generates a "report.tsv" file that can be opened in Excel. This shows how many pixels have been affected in the non-linear process. The stable % number gives you an average of how far the QuickNII output was from the non-linear correction, considering the non-linear correction to be the best situation (100%).

	A	B	C	D	E
1	snr	name	segmented	changed	stable%
2	74	074-xxxxxxxx.png	33375	11672	65 %
3	76	076-xxxxxxxx.png	32595	7474	77 %
4	92	092-xxxxxxxx.png	43829	13132	70 %
5	122	122-xxxxxxxx.png	56770	20217	64 %
6	148	148-xxxxxxxx.png	53453	26350	50 %
7					

6. References

- Puchades MA, Csucs G, Ledergerber D, Leergaard TB, Bjaalie JG (2019) Spatial registration of serial microscopic brain images to three-dimensional reference atlases with the QuickNII tool. PLOS ONE 14(5): e0216796. https://doi.org/10.1371/journal.pone.0216796