



Alignment and Reconstruction

Kitware Course Week, Clifton Park, NY

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Outline

- Introduction
- Tilt series
- Aligning data
- Reconstructing data

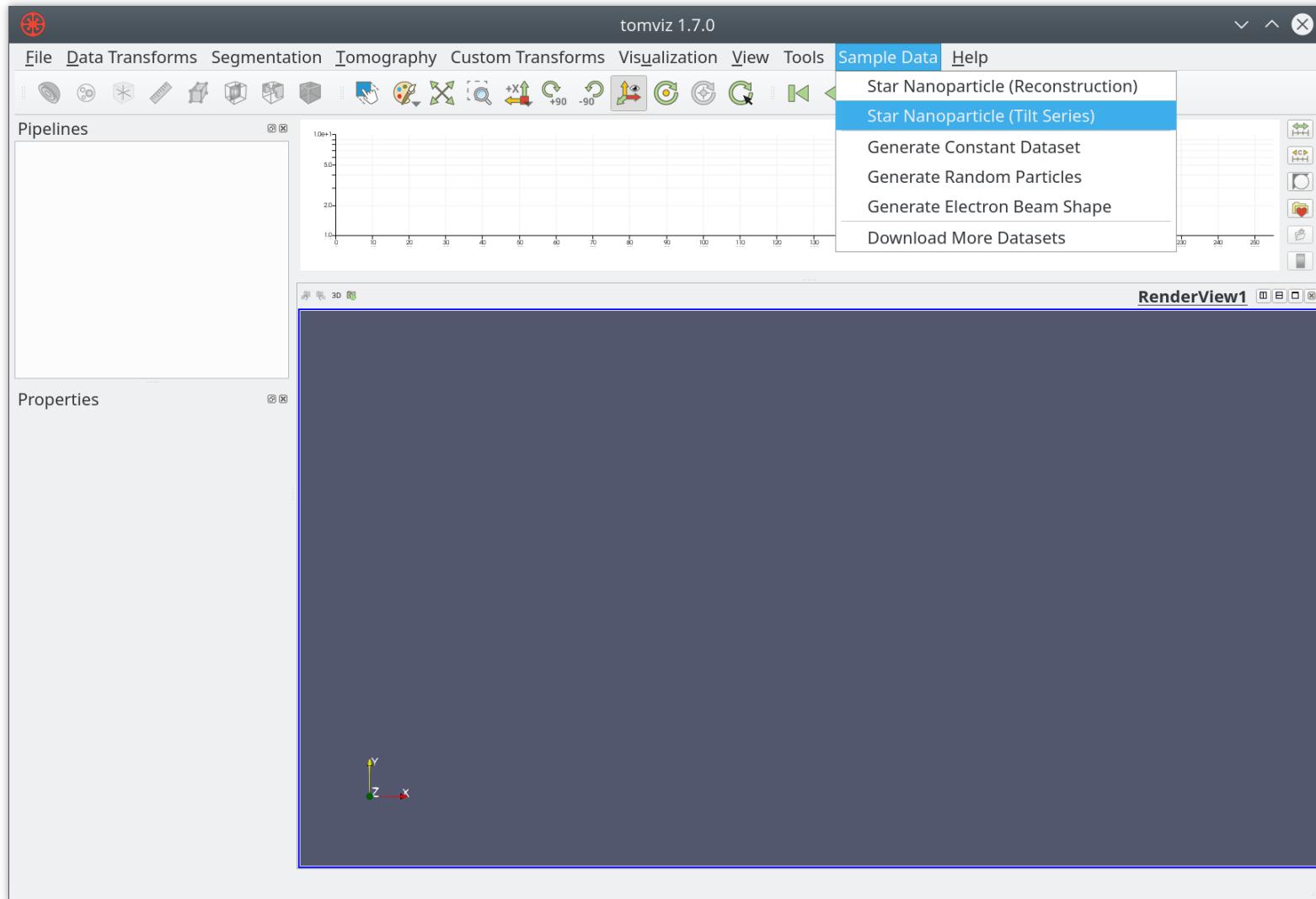
Introduction

- Two main types of data—volumes and tilt series
 - Volumes are from tomography or sectioning
 - Tilt series are projection series with angles
- Specialized tools for each kind of data
 - Tomography menu has tools for tilt series
 - Data Transforms and Segmentation menus for volumes
 - Add your own to Custom Transforms menu

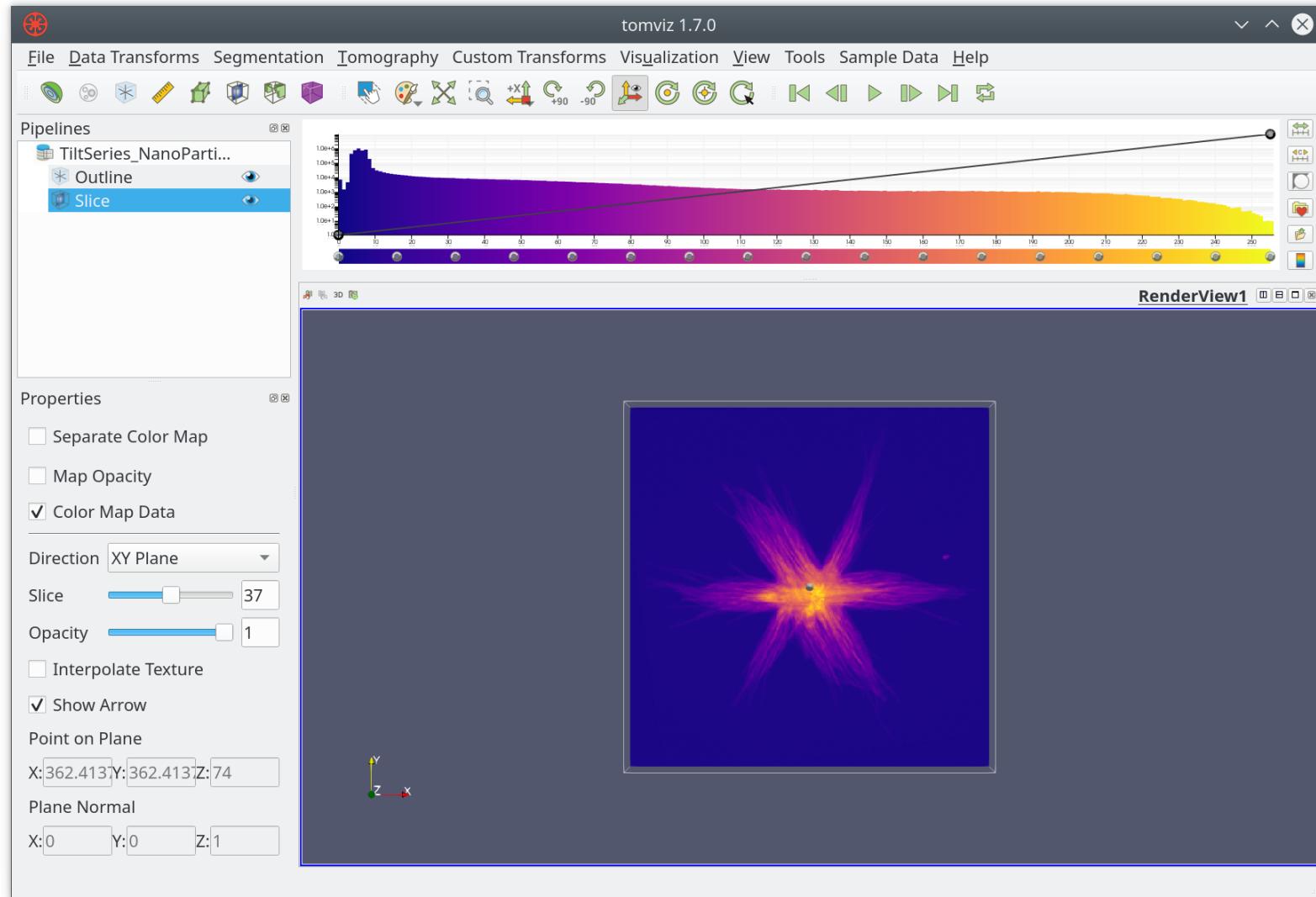
Tilt series

- Application comes bundled with a sample tilt series
 - It will automatically set the tilt angles
 - Normally this must be done manually
- Two major types of data that are loaded
 - Defaults to being treated as a reconstruction
 - Can be marked as a tilt series

Loading sample tilt series



Sample tilt series



Creating a tilt series

Tomography

Custom Transforms

Visualization

View

Mark Data As Volume

Mark Data As Tilt Series

Set Tilt Angles

Pre-processing:

Bin Tilt Images x2

Remove Bad Pixels

Gaussian Filter

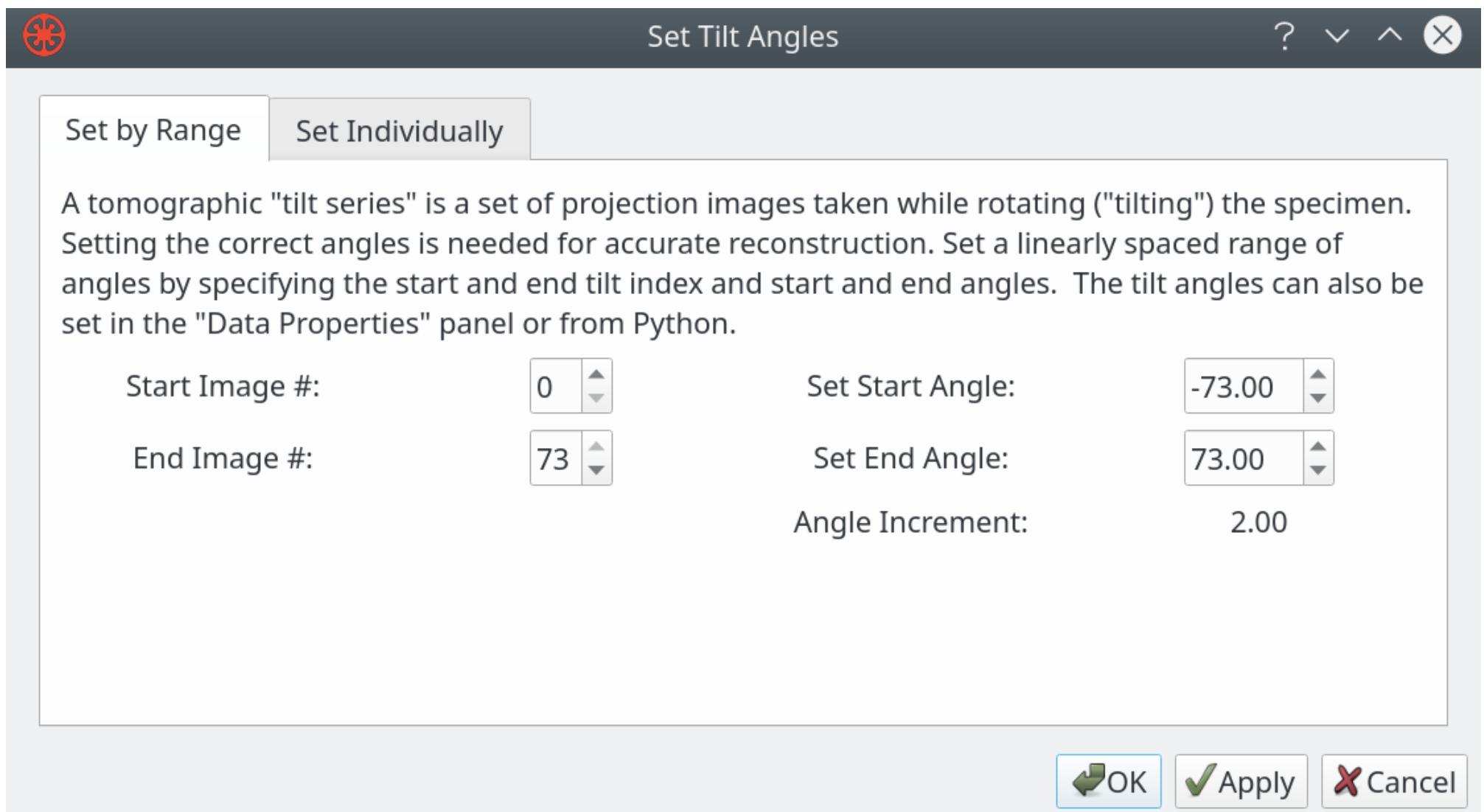
Background Subtraction (Auto)

Background Subtraction (Manual)

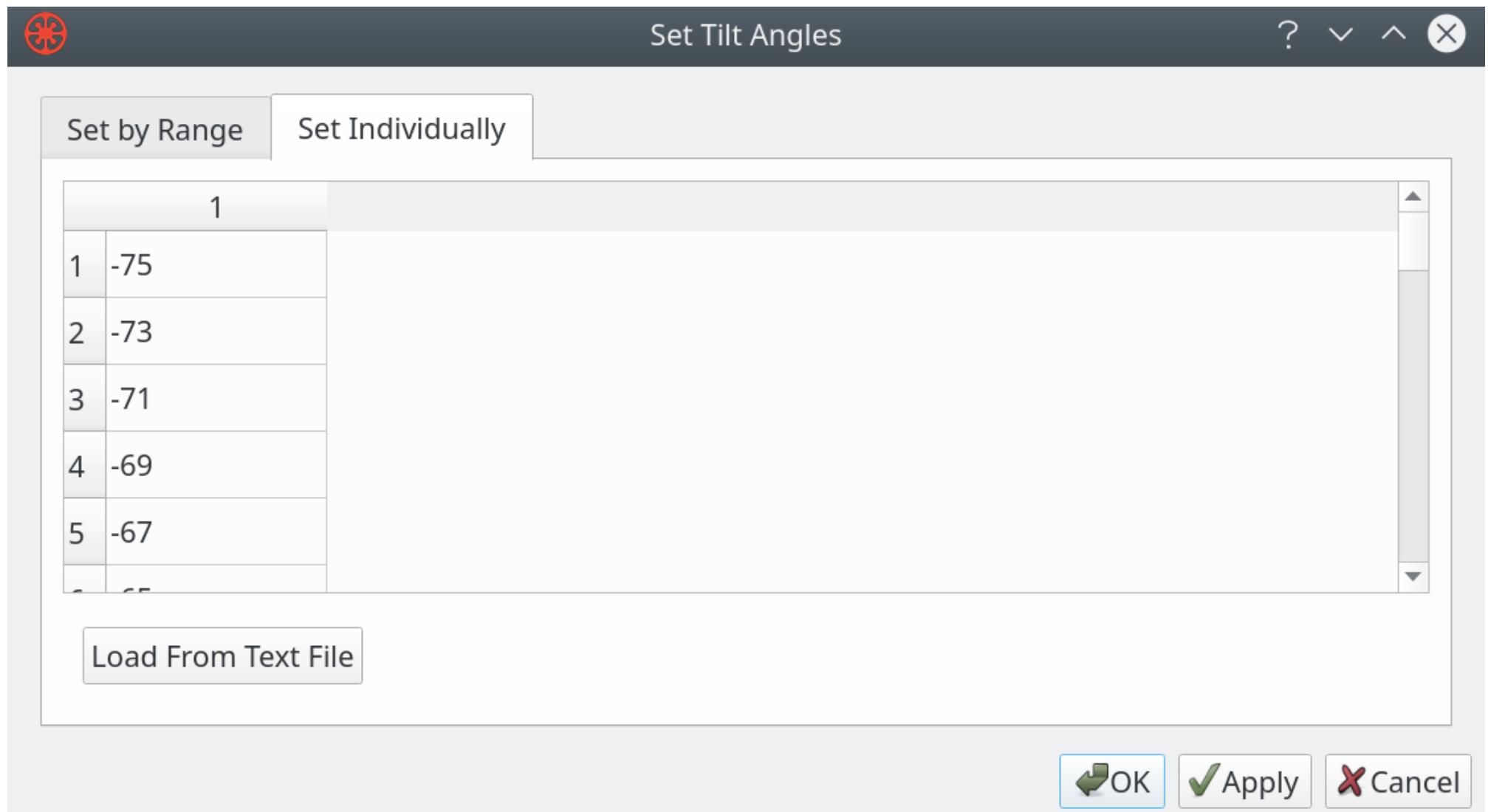
Normalize Average Image Intensity

2D Gradient Magnitude

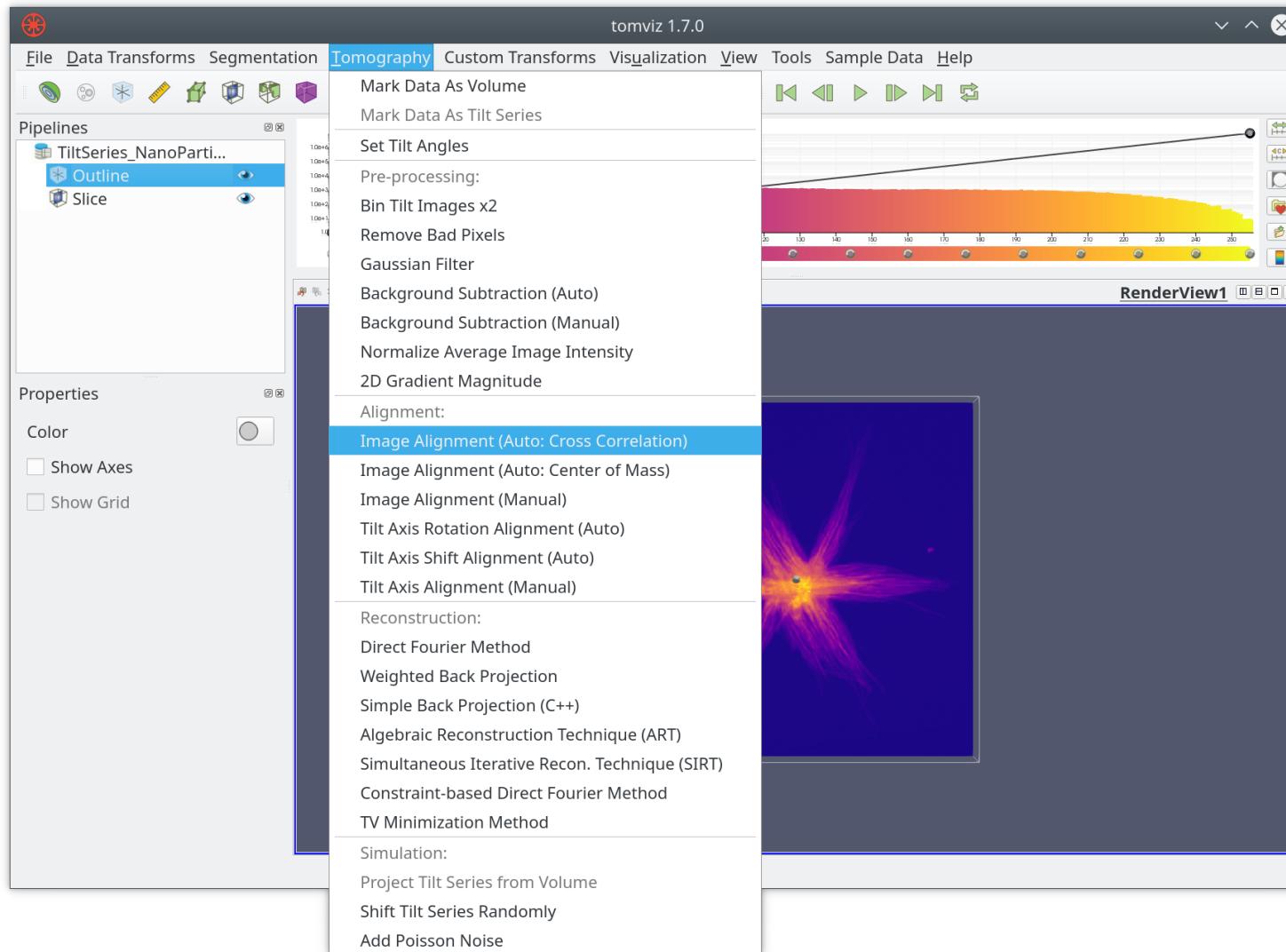
Set by range



Set individually



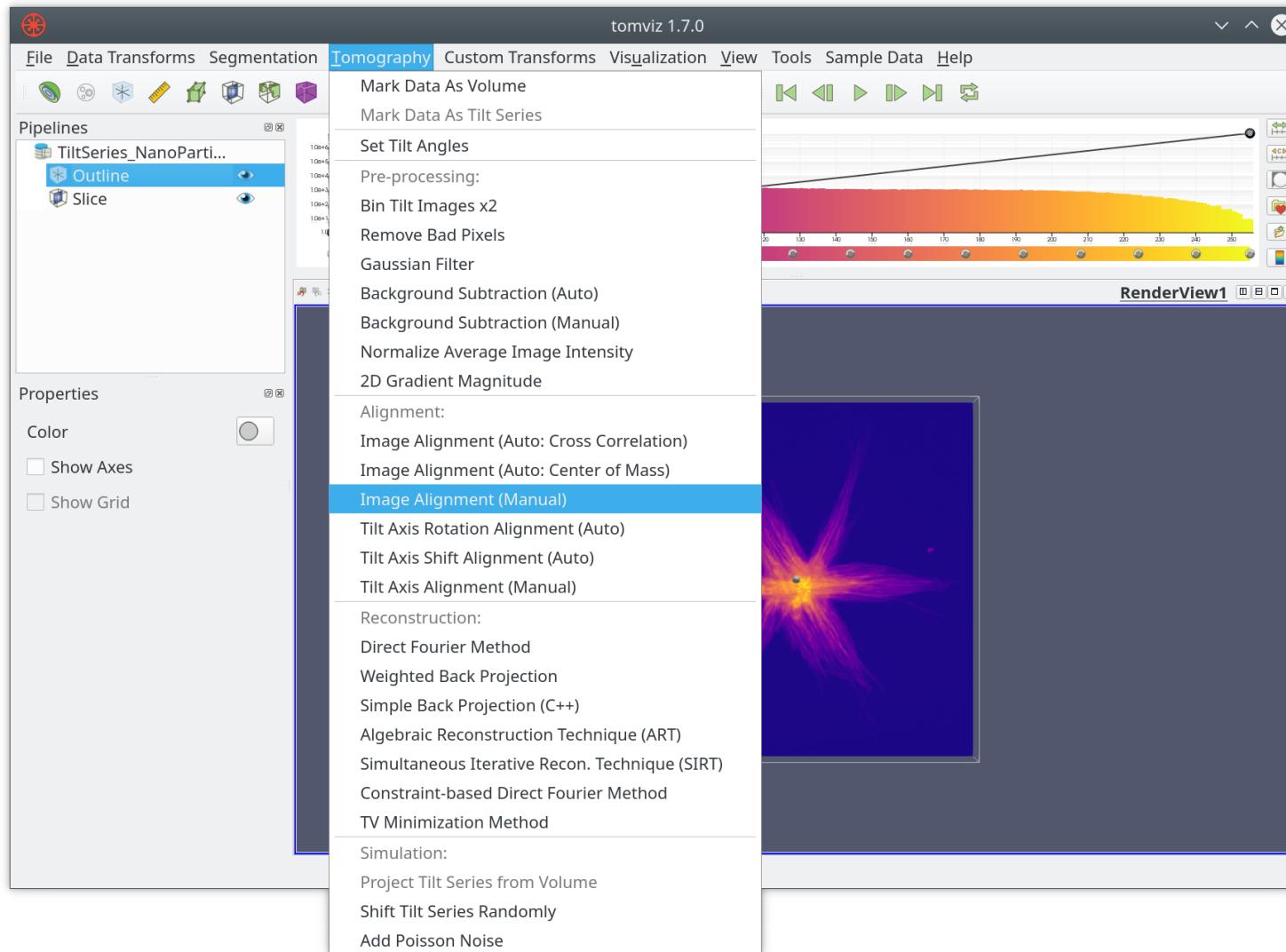
Aligning data



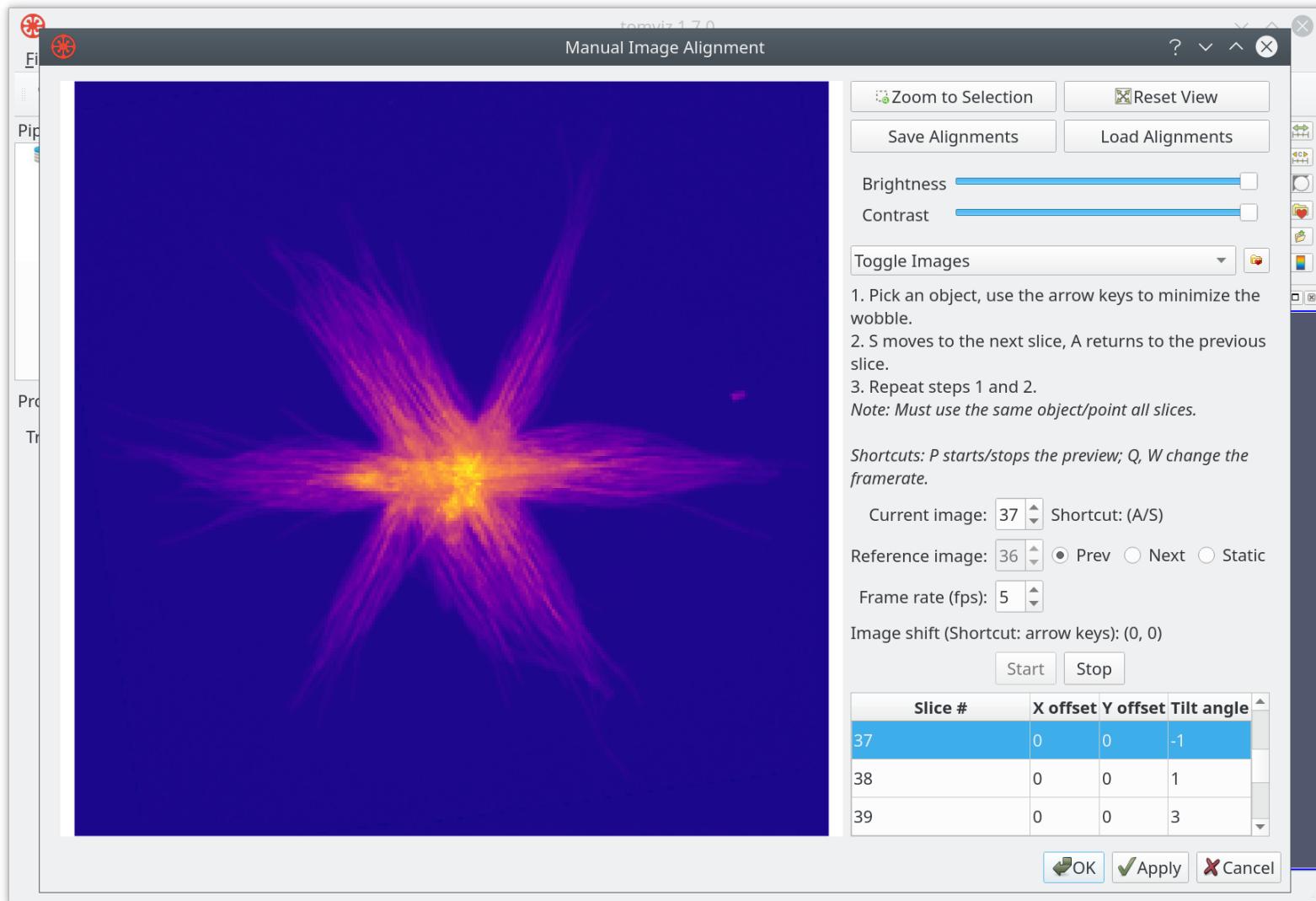
Manual alignment

- Some automated alignment algorithms are available
- Manual alignment of images offer a number of tools
 - Can be saved to a state file, and edited again
- Toggling between images, or image differencing
- Ideally use a fiducial marker to align
- Can align to previous, next, or to a fixed reference
- Keyboard shortcuts available to align, switch images
- Zoom, brightness, contrast available to aid alignment

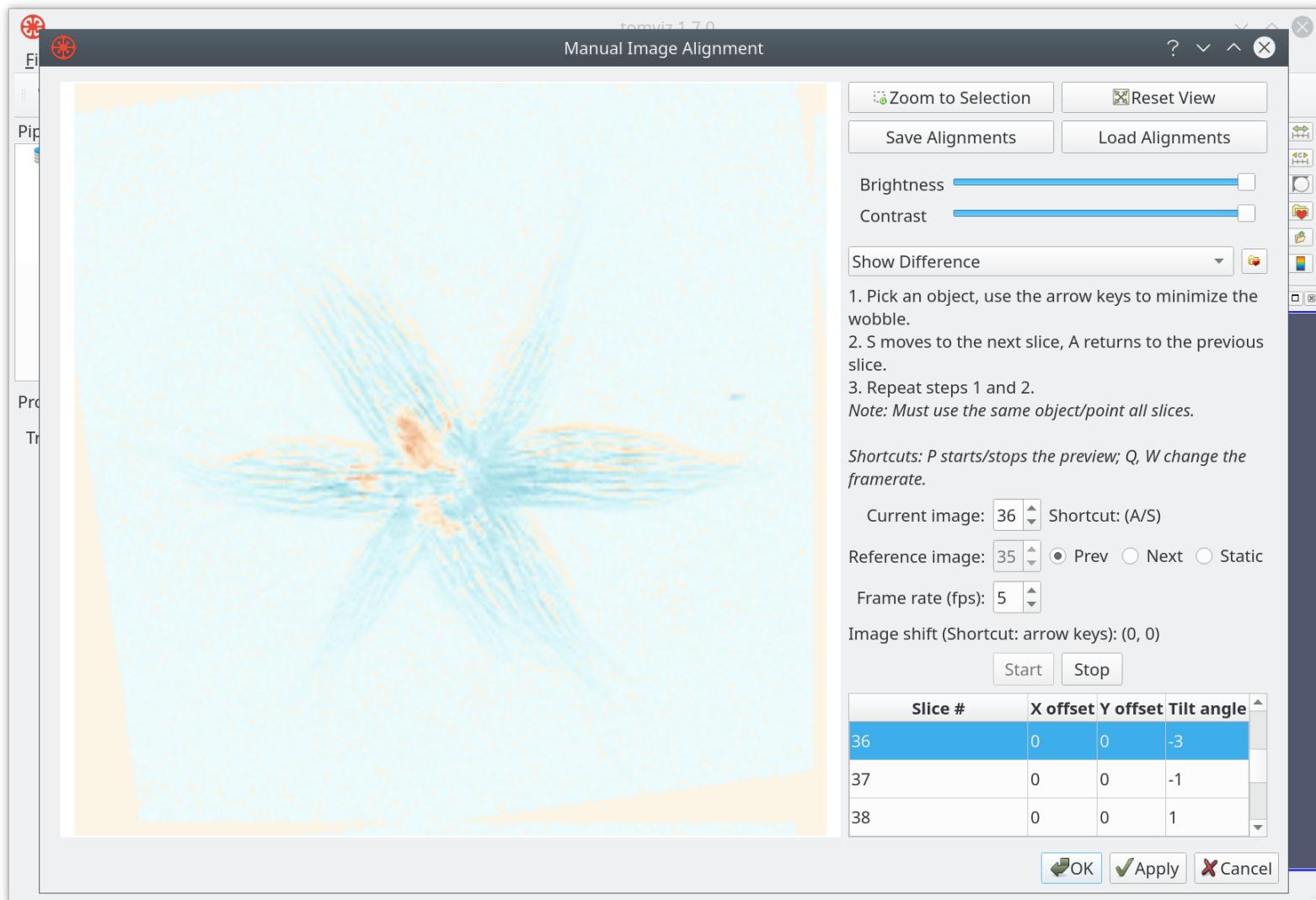
Manual alignment



Alignment dialog



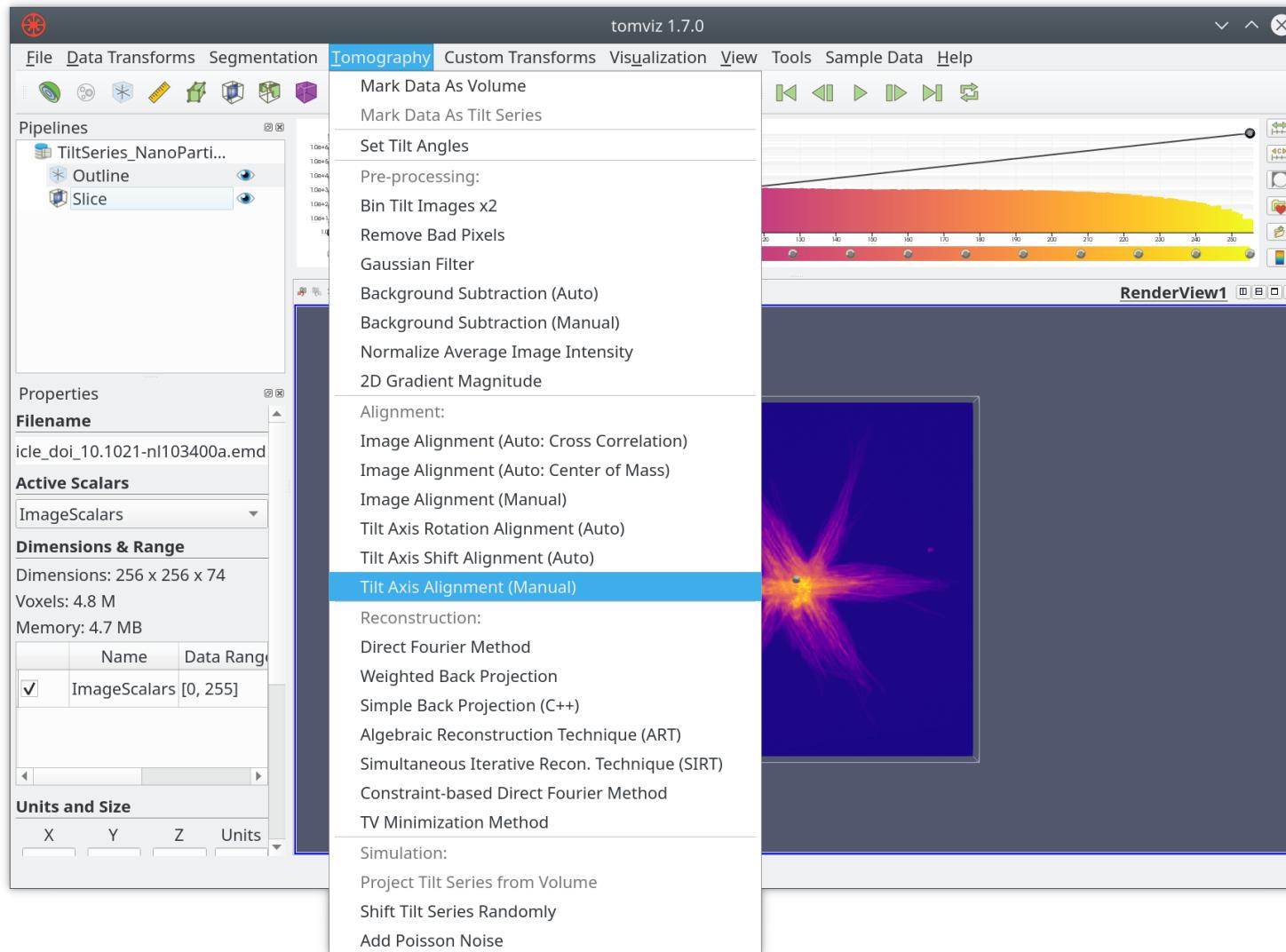
Manual alignment differencing



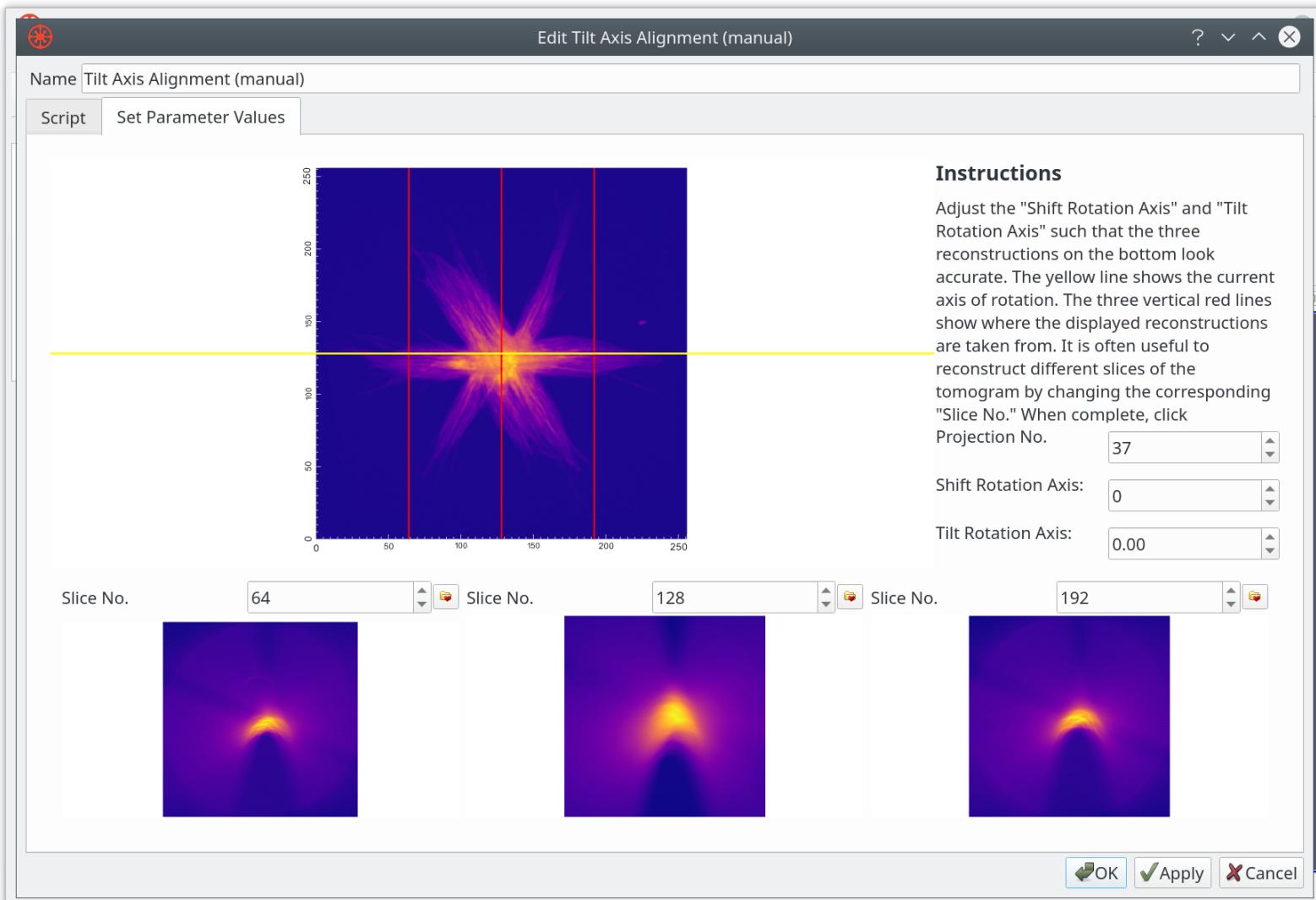
Tilt axis alignment

- Need to determine the tilt axis angle and position
 - This is in addition to aligning the images
- Dedicated dialog for manual determination
 - Some algorithms available to attempt fitting
- Choose three slices to show a quick reconstruction
 - Position one on a fiducial particle
 - Position the other two on interesting features
- Fiducial nanoparticles are ideally spherical
 - Crescent shaped when in the wrong position
- Color maps can be edited for each reconstruction

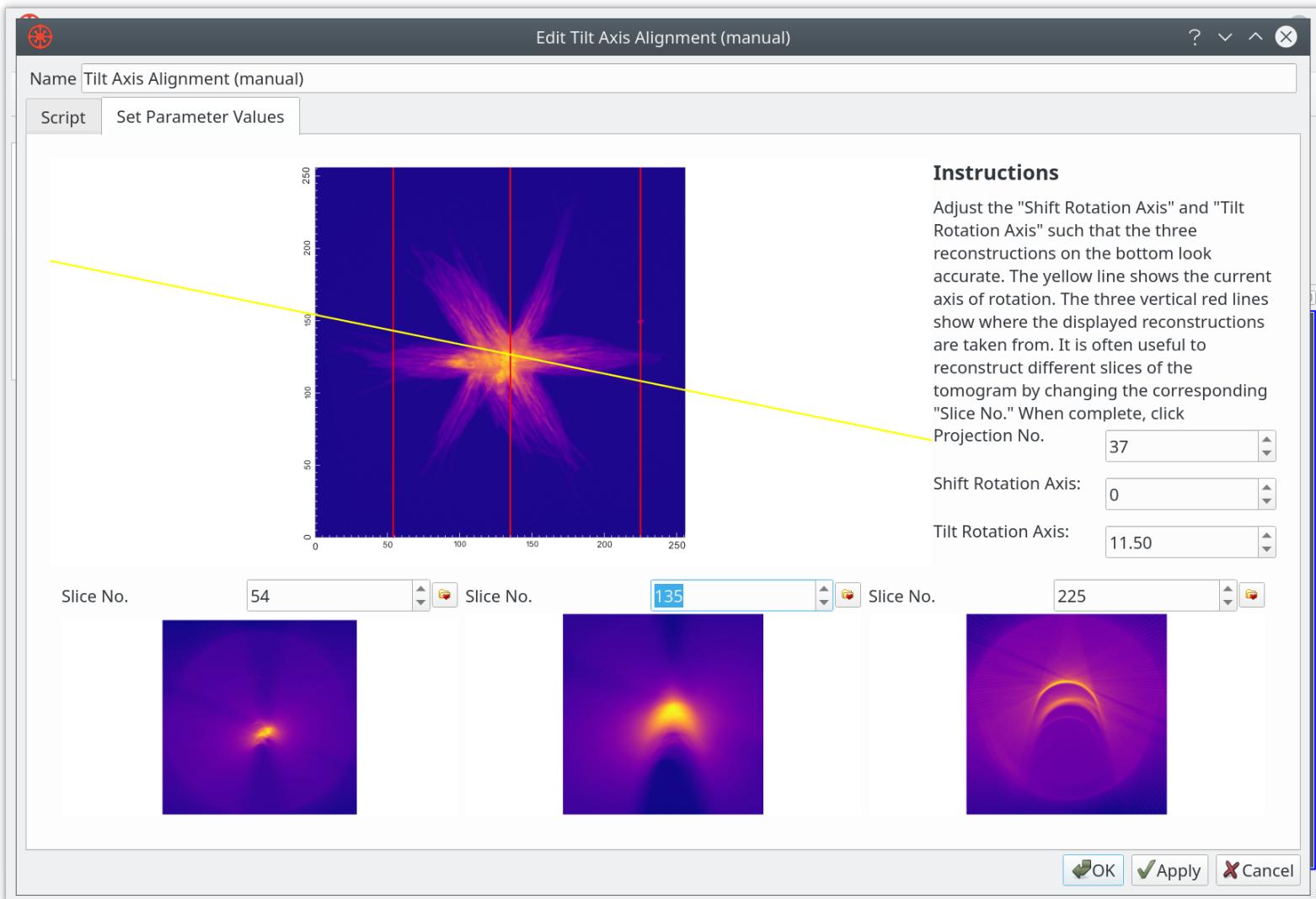
Tilt axis alignment



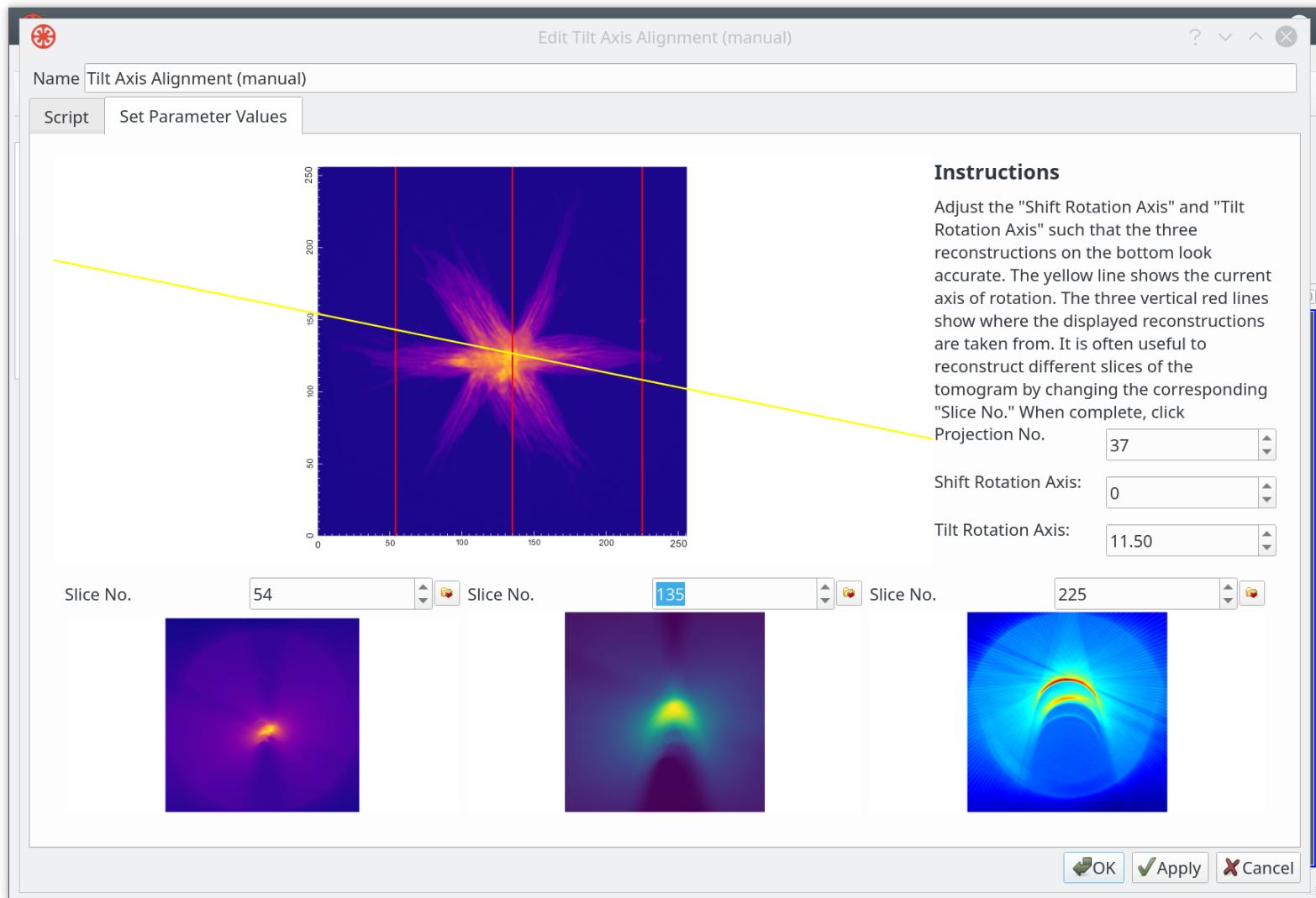
Tilt axis dialog



Tilt axis dialog



Tilt axis dialog

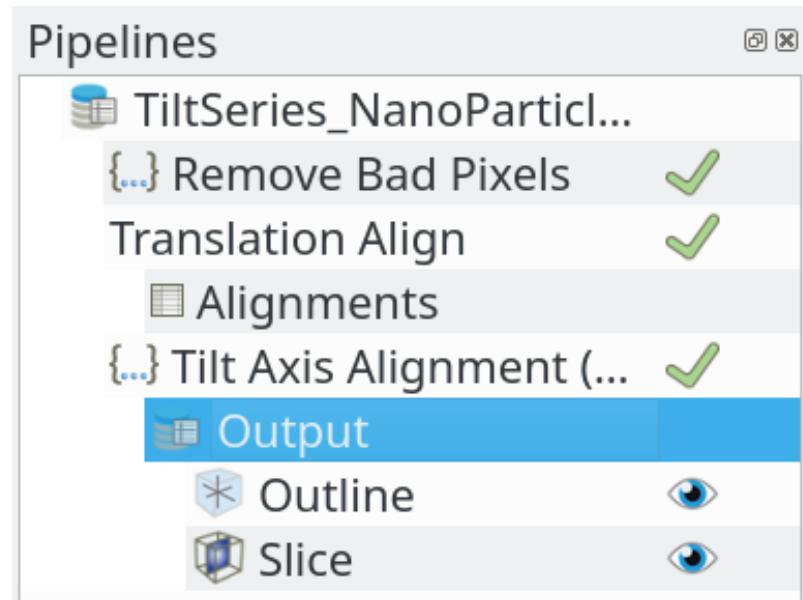


Reconstructing data

- A number of reconstruction techniques available
- This is the *most computationally intensive* step!
- Save your work often, consider downsampling it
- Some available algorithms include
 - Simple back projection
 - Direct Fourier
 - Weighted back projection
 - Many others, and it is easy to add more
- Most in Python, can be used as starting points

Pre-reconstruction pipeline

- Typical pipeline before reconstruction
- Some pre-processing, image and tilt axis alignment



Reconstruction menu

- Available reconstructions in the tomography menu
- Most are developed in Python, one in C++

Reconstruction:

Direct Fourier Method

Weighted Back Projection

Simple Back Projection (C++)

Algebraic Reconstruction Technique (ART)

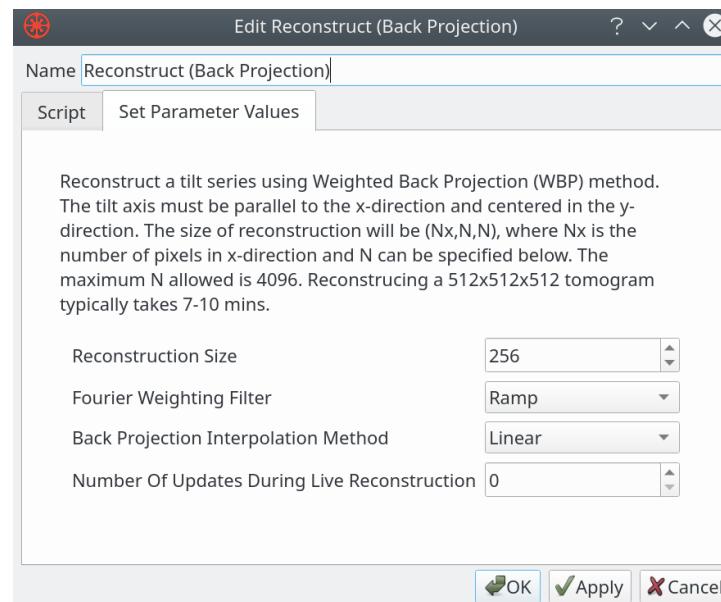
Simultaneous Iterative Recon. Technique (SIRT)

Constraint-based Direct Fourier Method

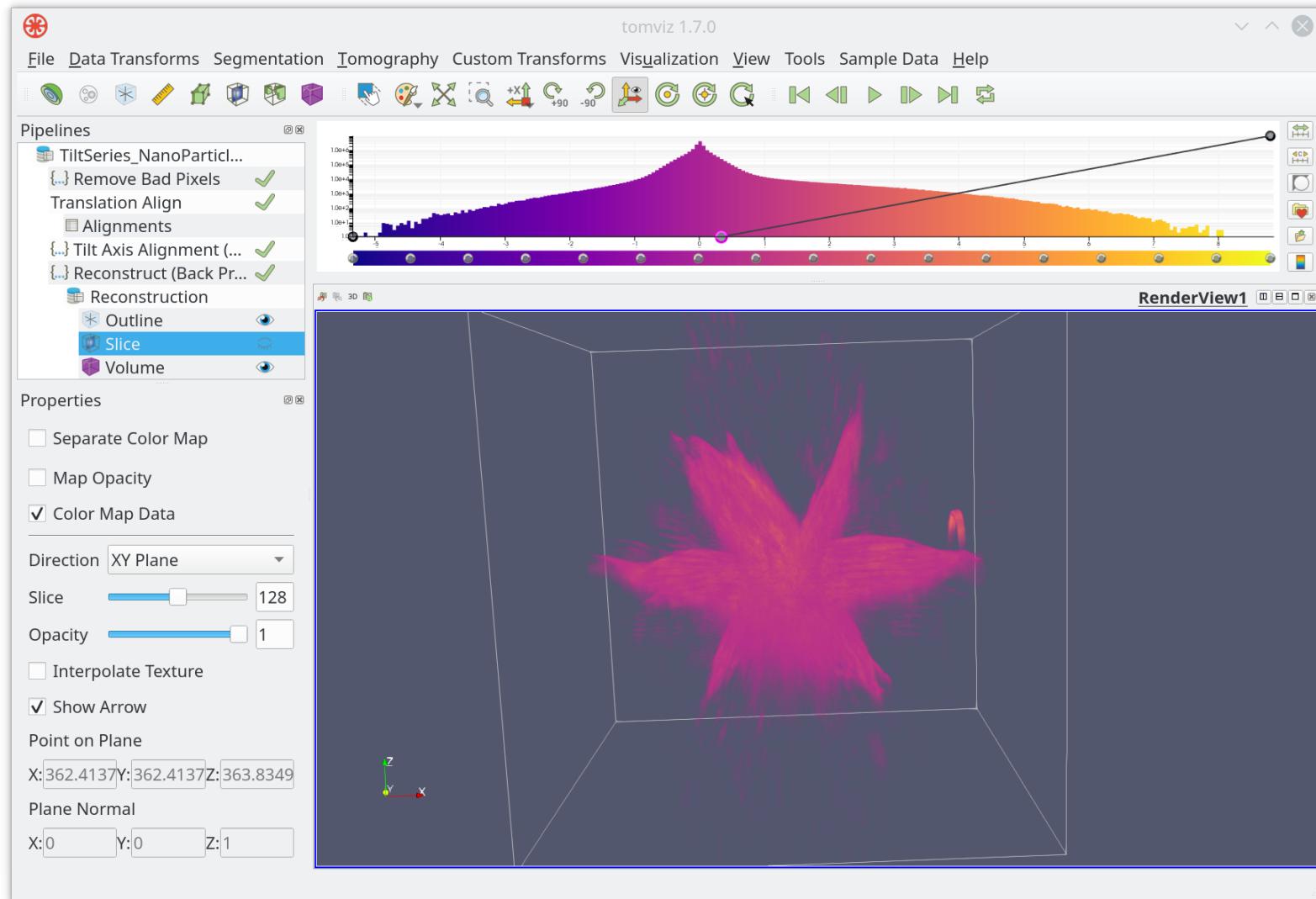
TV Minimization Method

Weighted back projection

- Enter the parameters shown
- Run the reconstruction

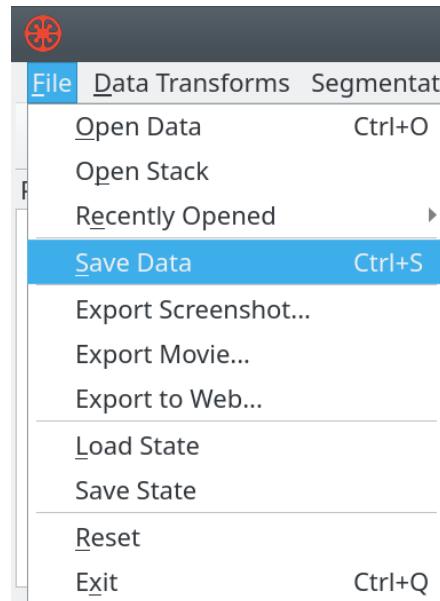


Weighted back projection



Save the reconstructed data

- Shown as a child data set
- Highlight it (by clicking on it)
- Go to the file menu, and click save data



Advanced reconstruction techniques

- All but one reconstruction algorithms—Python
 - You can examine the code in the application
 - You can even modify them and see the result
- Tomviz offers ready-to-use algorithms
 - It is also designed to enable others to add more
 - Experiment locally, consider making pull requests
- Modified operators are saved in the state file

Summary

- Gone from importing a tilt series
- Looked at pre-processing, alignment
- Reconstruction of the data
- Saving pipelines and data

