

aXis 2000

Analysis of X-ray microscopy Images and Spectra (02 Jan 2023)

aXis2000 - **Analysis of X-ray microscopy Images and Spectra** - is an [IDL widget](#) for viewing, comparing and processing X-ray microscopy images, spectra and spectromicroscopy (combined image & spectra data). IDL stands for [Interactive Data Language](#), a scientific computing platform developed by Research Systems Inc (RSI), currently owned by [L3Harris](#). aXis2000 contains scripts developed by Chris Jacobsen, Carl Zimba, Adam Hitchcock and others. The widget platform was written by Adam & Peter Hitchcock. It is maintained and frequently updated by Adam Hitchcock. It can be obtained from unicorn.mcmaster.ca/aXis2000. It operates on Windows (WIN), Unix (X) and Macintosh (MAC) versions of IDL.



A compiled version (**aXis2000.sav**) for use with [IDL Virtual Machine](#) is available. This allows access to the power of aXis2000 without needing to purchase an IDL license. Please note that there are some features of aXis2000 that work with the licensed version, but not with the VM version, and that the specific details of these problems depend on the versions of both IDL and aXis2000. I would appreciate it if you would notify me by email (aph@mcmaster.ca) about problems with the code or if you wish to make suggestions for improvements. If you make extensions or corrections to the distributed source files, or develop new IDL routines for specific data processing functionality, I would appreciate receiving a copy of your code revisions with sample data, so I can evaluate and incorporate in future versions.

*I thank all the people who have written scripts that went into this, including my son, **Peter Hitchcock** who helped set up the basic widget structure; **Carl Zimba** (Photons Unlimited) who supplied ZSTACK and extensively improved the overall package in 2000; **Eli Rotenberg**, **Jonathan Denlinger**, **Stefano Cerasari**, **Tolek Tyliczszak**, **Andy Smith**, **Andreas Scholl**, **Göran Johansson**, **Jacob Stewart Ornstein**, and many others. SPECIAL thanks to **Chris Jacobsen** (Stony Brook, nsls) for sharing his STACK_ANALYZE and PCA_GUI codes, **Rick Kneedler**, for providing the basis for the stack-fit routine, and **Billy Loo** for providing the SF X-ray spectral predictor based on, the Henke mass absorption coefficients, and the Conjugate Gradient Optimization routine (ax_cgo).*

TO START aXis2000: after [installing aXis2000](#) (see end of this file) on Windows, Unix or Mac OS: Start **IDL** ;

If you have set the Preferences (in IDL) so that **axis2000_batch.pro** is the start file, aXis2000 will launch automatically.

Otherwise, type **axis2000** on the IDL command line.

If you quit aXis2000 and stay in IDL, you can restart by typing **axis2000**

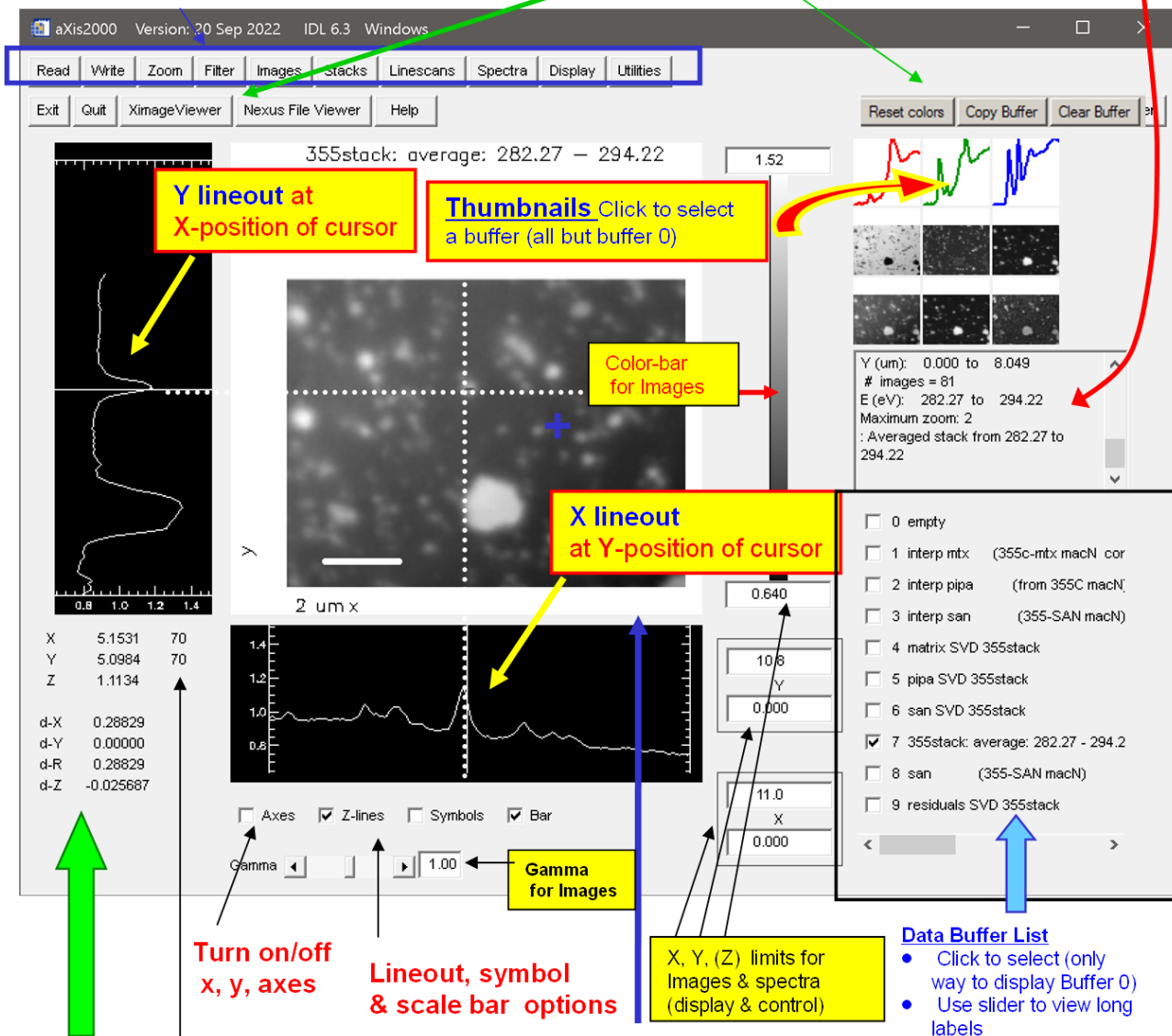
Jan2023

Features of the aXis2000 widget

aXis2000
Messages,
Hints and log

Pull-down menus

single action menus



Cursors

(X,Y,Z) – at cursor pixel indices
(dX,dY,dZ) – change over line (images) or between cursors (spectra)
dR – distance along line (images only)

Main Image

- Displays currently selected image or spectrum (or group of spectra, if *display~overplot* ; or group of images, if *display~thumbnails* is used)
- Size of AXIS2000 can be adjusted from 0.5 to 2.0 of its nominal size (360x360 pixels in Main Image) by the size parameter in *axis.ini*

Mouse

- First click – cursor and lineout; arms line generator
- Second click – draws and documents line (image) ;
- reports difference in cursors (spectra)
- Third click – clears line and cursor information

Notation conventions used in this reference manual for aXis2000

BOLD indicates a pull-down menu command

TOP LEVEL items are bold, underlined, and light blue highlighted

Utilities~print~annotated is a third level pull-down menu under **Utilities** first level button

Linescans indicates an internal bookmark or a hyperlink to a webpage.

Hard copy: Printing the Main Image is achieved by using *Utilities~print*. aXis2000 prints via the IDL Printer virtual device. S

Default parameters: The user can define a number of properties of the aXis2000 widget by editing the axis initialization file (**axis_win.ini**, **aximacos.ini** or **axis_unix.ini**).

I find it convenient to document data analysis carried out with aXis2000 by taking snapshots of the display [**Snipping Tool** in Windows10 or **MWsnap** (freeware, <http://www.mirekw.com/>), and transferring via the clipboard to **Powerpoint**, **Keynote** or similar program.

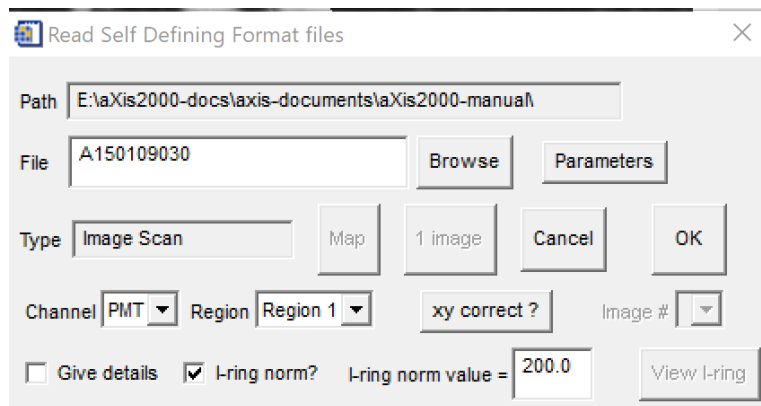
In Windows 10 I use the free, open source program, [paint.net](http://www.paint.net) , to modify snapshots from aXis2000.

TOP ROW PULL DOWN MENUS (Click on the menu name to go to that part of the manual)

Read Write Zoom Filter Images Stacks Linescans Spectra Display Utilities

READ for all types of data ([stacks](#), images, linescans, spectra, ROIs etc)

STXM (sdf) - [self defining files](#) containing all types of data from the ALS, SLS, Bessy and CLS STXMs are read by the following [widget](#). (single images; multi-region images, single spectra, multiple point spectra, linescans, image sequences for one region or multiple regions; DAQ, all types of motor scans, etc)



STXM (sdf)
STXM (NeXus)
PEEM (lox)
Images >
Spectra >
Stacks >

All data written by the ALS, CLS, UVSOR STXMs are described by a self-defining header file (*.hdr) which contains the information needed to identify the type of data contained in the associated data files. After selecting a file, the widget indicated above can be used to :

Browse – to select a file (you can also edit the file name– hit **ENTER** after changing)

Parameters - list the header, which contains microscope, beamline & source parameters

Map – generates a component map from the first 2 images of a stack [OD(2)-OD(1)]

To do this from any 2 images in a >2 image stack use *stacks~analyze~stack process*

1 image – (only for stacks) read in one image from a stack (select using **Image#**)

Cancel – dismiss the STXM (sdf) read in widget

OK – read the file, using the selected options

Channel - select data channel from pull down list, generated from header

Region - select spatial region (area or point) from pull down list, generated from header

xy correct ? - if checked, and the data was recorded, shifts position of each pixel to that reported by the interferometer guidance system

Image# - select specific image of multi-image stack

Give details – plot I-ring values for a stack

I-ring norm? – if checked, all data intensities are adjusted as if Ring current is the value given in I-ring norm value

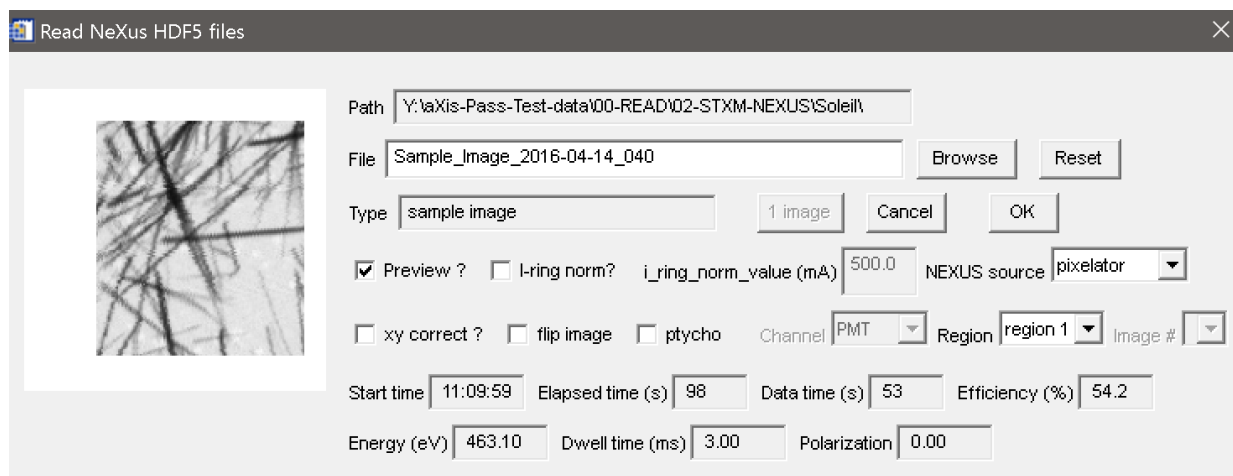
I-ring norm value – CLS – 200 mA (default); ALS – 500 mA

View I-ring – plot I-ring values for a stack

Selecting **OK** then reads the information in the associated image (*.xim) or spectral (*.xsp) ascii files into one or more aXis2000 buffers. Image sequences are converted directly to *.ncb files

READ (continued)

STXM (NeXus)



Browse - to select a file

Reset – clears Path, File, Type

image – (only for stacks) read in one image from a stack (select using **Image#**)-1

Cancel – dismiss the STXM (sdf) read in widget

OK – read the file, using the selected options

Preview – if set, display NeXus data

I-ring norm ? – if checked, all data intensities are adjusted as if ring current is defined value

I-ring norm value CLS (cryo-STXM) – 200 mA; Soleil – 500 mA

NEXUS source pixelator (SLS, Bessy, Soleil, Diamond) or pySTXM (CLS cryo)

xy correct ? - if checked, and the data was recorded, shifts position of each pixel to that reported by the interferometer guidance system (experimental)

flip image – required for some versions of pySTXM image files

ptycho - read in STXM type images generated when measuring in ptychography mode

Channel - select data channel from pull down list, generated from header

Region - select spatial region (area or point) from pull down list, generated from header

Image # - select specific image of multi-image stack

Start time - clock time at start of acquisition

Elapsed time - time of acquisition

Data time – time predicted from dwell and line delays, assuming no other delays

Efficiency – ratio of data / elapsed time

Energy (eV) – photon energy

Dwell time (ms) – dwell time

Polarization – parameter for photon polarization (converted to LCP, RCO, Lin-H, lin-V and displayed on y-axis label)

READ (continued)

PEEM (Lox) – Reads data written by Lox, the data acquisition for the CLS PEEM (called CaPeRS = **C**anadian **P**hotoemission **R**esearch **S**pectromicroscope).

Lox data is described by a header file (*.lox) which contains the information needed to identify the type of data contained in the associated data files, along with all data acquisition and PEEM parameters. After selecting a file, the Read Lox widget can be used to :

Browse – to select the file (or edit the filename box)

Parameters - list the header, which contains all microscope parameters

Data Channel - select from pull down list, generated from header

Region - select from pull down list, generated from header

I-ring norm? – if checked, all data intensities are adjusted as if Ring current is 500 mA

Image Energy - select specific image of multi-image NEXAFS Image scan

Map – (only for a stack) – convert a 2-imagestack into an OD difference map

1 image – (only for NEXAFS Image Scan) read only the image identified in the image energy pull down list (which displays the photon energy)

Image Energy (eV) - list of energies of all the images in a stack

Dark Level – set camera dark level

Field of View - set field of view of the image (microns)

Cancel – dismiss the ALS STXM read in widget

Selecting **OK** then reads the information in the associated binary image (*.tif) or ascii spectral (*.lox) files into one or more aXis2000 buffers. Image sequences are converted directly to the aXis2000 binary format (*.ncb).

READ (continued)

Images

AXIS – images written in aXis2000 **binary** format (*.axb)

This is useful for saving derived results. It is the default output format, and the required input for some routines

ALS-xyt – file format for pattern generator (STXM)

Bessy – read 1 image or a stack from files of old Bessy-I STXM

Bessy	>	STXM (old)
Graphics	>	TXM

Graphics - standard formats (**BMP**, **GIF**, **JPG**, **PNG**, **TIF**).

TIF format handles 1-channel & 3-channel (RGB) formats

For each image format one can read the image as

image (un-modifiable, preserves colors)

or as **data** (modifiable, pixel indices as (x,y)-values)

CAUTION: there are known bugs in these routines,
with some inversions and strange color distortions

AXIS	
ALS-xyt	
Bessy	>
Graphics	>
NSLS	>
PEEM	>
Ptychography	>
ROI	>
SPEM	>
Text	>
Twinmic	>
TXRM	>
XRF	>
Other	>

Graphics	>	BMP	>	
		GIF	>	
		JPG	>	
		PNG	>	
		TIF	>	image data

NSLS – read images written by NSLS I STXMs (pre 2012 or so)

old (*.nc) – prior to ~2002 (NetCDF)

stxmIV – last used data format

cryo - format from cryo-STXM

old (*.nc)
stxmIV
cryo

READ~Images (continued)

PEEM

ALS-PEEM2 – reads in 12-bit and 16-bit ALS-specific formats with a **widget**.

(NB the **Elmitec** read command uses the same structure to define image and image sequence parameters)

NB – this routine may not work in the Virtual Machine (*.sav) version

ALS-PEEM2

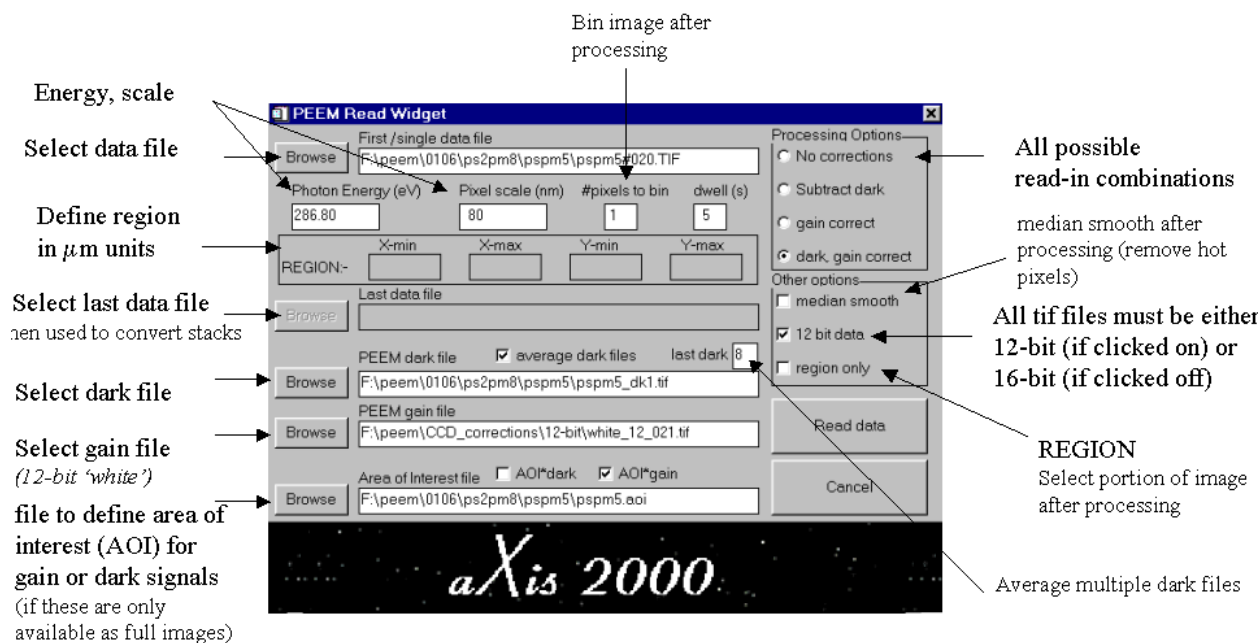
ALS-PEEM3

Elmitec

Lox-PEEM

NSRRC

Mephisto



ALS-PEEM3 – reads data format for PEEM3 (implemented Jan-2007, Andreas Scholl)

Elmitec - read PEEM files.

- **dat** – read in 16-bit (12-bit encoded) files written by UView2002
- **tif** – read in 8-bit (3-channel grayscale) exported by UView2002

Lox-PEEM – read in 16-bit (12-bit encoded) files written by UView2002

NSRRC – PEEM

Mephisto – read PEEM data from Mephisto (old binary 512x512 from a CCD)

ALS-PEEM2

ALS-PEEM3

Elmitec

Lox-PEEM

NSRRC

Mephisto

dat

tif

READ~Images (continued)

Ptychography

Ptychography	>	11.0.2-display
ROI	>	Soleil
SGM_Image	>	Shapiro

11.0.2 display – animation of ptycho acquisition

Soleil tif file generated by Soleil ptycho acquisition (2019 version) – superseded by *.hdf5 file

Shapiro - output of Nov-2012 ptychography image analysis

ROI – region of interest definitions for

Lox – ROI for Lox format PEEM

Pem2 – ROI (=AOI) for pem2 data

aXis2000 stack – ROI for stack region selection (CJJ, CZ)

lox
pem2
aXis2000 stack

SPEM

ALS-SPEM – read in images recorded in XPS or NEXAFS mode using the ALS SPEM

NSRRC SPEM read in images recorded with Taiwan PEEM

ALS-SPEM
NSRRC SPEM

Text - read in images written as an array of Ascii (ie readable with a text editor like Notepad) , with or without standard aXis2000 header

no_header
with_header

Twinmic - read in images from Twinmic (Elettra) STXM

Transmission

Fluorescence

Twinmic	>	transmission
TXRM	>	fluorescence

TXRM – read in images written by X-radia software

Alba.Mistral

Xradia [STXM at Shanghai - stacks]

Alba.Mistral
Xradia

NB this only works on a Windows system & needs some special codes

(IDL-Java bridge) which are provided in the aXis2000 source package.

Contact Adam Hitchcock if you need instructions on how to set that up

XRF read in X-ray fluorescence (XRF) maps from different systems

APS – from GoeCARS XRF microprobe

Inca (Oxford) – from McMaster SEM (Tescan) running Xmax SDS

CLS SGM – from Amptek (spectral mode as an array)

Twinmic – XGLabs data from Twinmic

APS
Inca (Oxford)
CLS SGM
Twinmic

OTHER - mostly OLD formats

Ascii image (Reals or Integers)

ALS-STXM-7.0 image files (ascii) – data channels:

- im0 = OSA signal (Io);

- im1 = transmitted signal;

- im2 = other (e.g TEY, luminescence, etc)

ALS-STXM-7.0 linescan – linescan files from ALS-STXM7.0.1

ALS-XM1 – data files from full field microscope at the ALS (rudimentary)

Ascii image	>	reals
ALS-STXM-7.0		integers
ALS-STXM-7.0.linescan		
ALS-XM1		

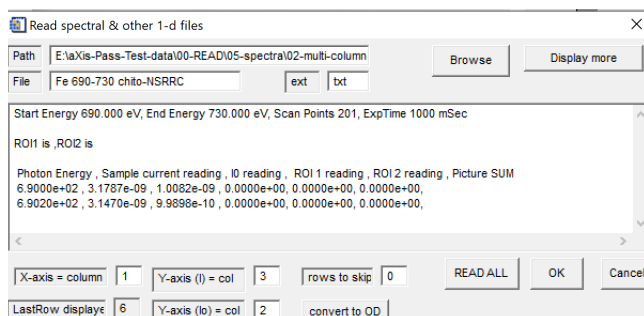
READ~Spectra

AXIS – read spectra from ascii format files (*.txt)

This can be ascii 1-d data written by aXis2000 or many generic formats of multi-column data with or without headers. (default ext. = *.txt)

alignment – alignment (x,y – shifts) from Zimba, or Jacobsen aligners

multi-column – guided read-in of any multi-column, header ascii data file, with ability to skip lines, choose columns, multi-column read-in, etc



AXIS
alignment
multi-column
ALS-PEEM
CLS-SGM >
Lox-PEEM
MaxIV_SoftiMax_h5
MSA
NSLS >
OLD >
Twinmic >
XAS >
XRF >

Browse – select file

Display more – add another 6 lines to the display buffer (to get to end of headers)

X-axis column – select data column for x-axis

Y-axis column – select data column for y-axis

Rows to skip – use LastRow displayed to identify which line to start read in

Read all – read all columns in a multicolumn file (x, many y)

OK – read only the identified (x,y) columns

Cancel – abort widget

Y-axis(Io) – column for Io intensities

Convert to OD - use 2 columns to generate OD as $\ln(I_0/I)$

ALS-PEEM – spectra from ascii file written by ALS PEEM 2

CLS-SGM

csv – comma separated variables format

written from web-based CLS-SGM converter software

[<https://sgmdata.lightsource.ca/users/>]

csv
hdf5

hdf5 – hierarchical data format version 5 – SGM TEY/ transmission and XRF-yield data as stored by the acquisition software. NB this current routine (May 2022) only reads the first entry.

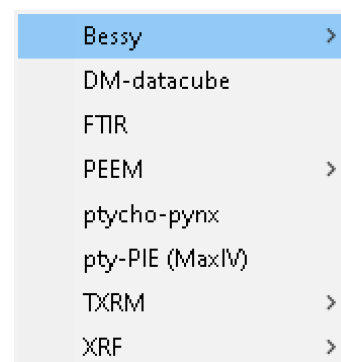
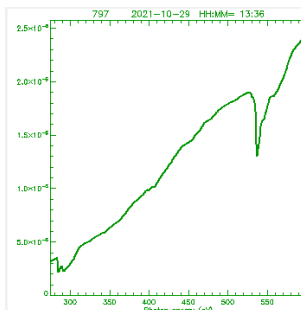
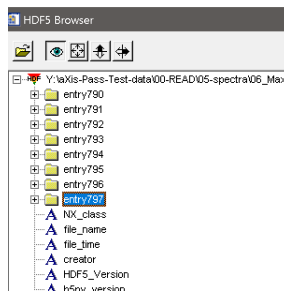
Lox-PEEM – read in multi-column spectra written by Lox (*.lox files)

READ~Spectra *(continued)*

MaxIV_SoftiMax_h5 – read in spectra or beamline monitoring data files written in hdf5 format

e.g. 20211029.h5 (just click on a single entry# of the multi-entry hdf5 file)

STXM (old)
TXM



MSA - read in Digital Micrograph spectra (TEM-EELS)

NSLS – read spectral data files in formats used at X1A

- **nc** - as-recorded (BSIF binary)
- **stxmIV** – stxm IV format (nc, but different than old-stxm)
- **ascii** - converted spectral format (generated by an X1A nsls utility)
- **map** - mapper style files (e.g. stack alignment shifts)

nc
stxmIV
ascii
map

XAS – (**X**-ray **A**bsorption **S**pectra) format with user settable header set up for bibliography (used for reference spectra, ZSTACK, NSLS, PCA_GUI and COREX bibliography)

single spectrum or multiple spectra written to common energy sampling (x-axis)

Amptek (*.mca)
Inca (Oxford)
XGLabs-text
XGLabs-hdf5

XRF

- **Amptek (*.mca)** – XRF spectra written from the Amptek software
- **Inca (Oxford)** – XRF spectra written from Inca (SEM, TEM)
- **XGLabs (*.dta)** – XRF spectra written from XGLabs software
- **XGLabs-hdf5** – XRF spectra written in hdf5 format from XGLabs software

Read~Stacks

Bessy

STXM (old) read stacks from Thieme / Gottingen scanning TXM

TXM read stacks from cryo-TXM

READ~Spectra *(continued)*

DM-datacube – converts binary data exported from Digital Micrograph image sequences into a aXis2000 *.ncb stack

FTIR – converts Nicolet Thermo (ver 7) map files (sets of spectra in spatial array) to *.ncb stack (for Soleil FTIR)

PEEM

NSRRC - PEEM stacks from Taiwan

Soleil – NeXus format stacks written from Soleil PEEM

only versions of IDL above 7.0 can read Soleil NeXus (.nxs) files*

All other – converts PEEM stack data from various instruments (Lox, Sphinx, ALS-PEEM2, ALS-PEEM3, ALS-pre-Sep02 (peem2 old format), Other).

See BELOW: [STACKS~convert format ~ PEEM](#) for view of widget and full description)

NSRRC
Soleil
all other

Pytcho – PYNX –(Soleil) reads in selected type of tif files (object or probe, amplitude or phase) from sets of folders (1 / energy) OR all-from-1-folder

Pty-PIE (MaxIV) - reads in multiple reconstructions as sequence of *.txt files

([spectro-ptychography at MaxIV SoftiMax under development as of Jan 2023](#))

Alba.Mistral
Xradia

TXRM – stacks from full-field microscopes at Alba and SSRF

Alba-Mistral

Xradia (SSRF, Shanghai)

XRF – read in stacks of X-ray fluorescence images

Inca
STXM_control ▶
XGLabs ▶

Inca – converts X-ray fluorescence (XRF) maps written from the Bruker Inca raw export into a *.ncb stack

STXM control

one energy - converts XRF-maps measured at a single photon energy using STXM_control to a *.ncb stack

multiple energies - converts XRF-maps measured at multiple photon energies using STXM_control (stack-of-stacks) to a *.ncb stack with summing over user selected X-ray fluorescence energy range to make an XRF-yield stack

one energy
multiple energies

XGLabs – converts XRF-maps written by the Labview software controlling the Elettra Twinmic STXM to a *.ncb stack starting from **text (*.dta)** or **hdf5** data files

text (*.dta)
hdf5

WRITE store results (images, spectra, etc) for later use or transfer to other programs.

AXIS - automatically selects file type depending on buffer contents

- spectra written with header in ascii (***.txt**)
- images written with header in Z-matrix binary format using IDL system-independent binary coding (***.axb**)



AXIS
Graphics >
ALS-xyt
ascii image >
NSLS-image (*.nc)
SDF format
XAS single/multiple

Graphics

GIF – writes current Main Image (with any annotation etc) as a GIF file

- **image** (exactly as seen on main screen of aXis2000)
- **data** (the image without axes or labels)

JPG – writes the current Main Image (with any annotation etc) as a JPG file

- **image** (exactly as seen on main screen of aXis 2000)
- **data** (the image without axes or labels)

PNG – writes the current Main Image (with any annotation etc) as a PNG file

- **image** (exactly as seen on main screen of aXis 2000)
- **data** (the image without axes or labels)

TIF - writes the current Main Image (with any annotation etc) to a TIF file.

- **image** (exactly as seen on main screen of aXis 2000)
- **data** (the image without axes or labels)



GIF ▶
JPG ▶
PNG ▶
TIF ▶

ALS-xyt – file with point-by-point (x,y,t) values to create patterns using **pattern_generation** function of STXM_control. NB after reading in a graphics image (tif, jpg etc), the *Images~Set XY Scale Utilities~change mesh* and *Spectra_gain* functions can be used to adjust size, point spacing (typically 50 nm) and intensities (0-1) in the *.xyt file this writes. At the STXM, the pattern_generation routine can then adjust exposure time through a multiplier.

ascii image – write image to array of **Real** or **Integers** for export to other programs (e.g. excel)

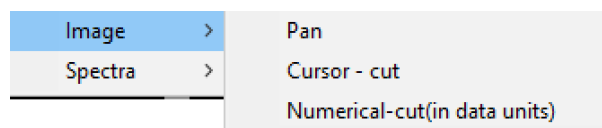
NSLS-image (*.nc) – writes NSLS [netCDF](#) image format (***.nc**) - used in stack_analyze

SDF format – write [self defining file](#) format [* .hdr, * .xim (* .xsp) files for images (spectra)]

XAS single/multiple - writes one or more spectra which are displayed in the main viewing window of axis2000 in XAS format with optional definition of detailed header. This is a useful way to transfer multi-spectral data for figures in a scientific plotting program like Sigma Plot, Origin etc

ZOOM (for images and spectra)

Image (NB –since expanding the scale is performed by cutting out the region of interest, the cursor-cut and numerical-cut commands can also be used to extract sub images)



Pan – pops-up a zoom window with a ~3x expansion

of the region around the cursor. Move the cursor on the main image to look at different areas. Left click to change zoom factor. Right click to end zoom. If the continuous lineouts option button at the bottom of the aXis2000 screen is OFF, then the zoomed image is only updated on each left mouse click.

Cursor – cut - stretchable box cursor used to define region. Data is cut from the displayed image and shifted to the working buffer (0).

Numerical – cut - numerical selection of range. Data is cut from the displayed image and shifted to the working buffer (0). The last values used to define the limits are saved and used the next time zoom~image_numerical is used, which allows accurate extraction of regions from many images (e.g. a set of component maps from fitting)

Spectra

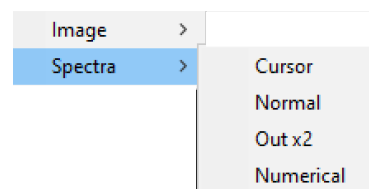
Cursor -Stretchable box cursor

Normal - return plot to full X-scale, same Y-scale

Out x2 - useful to "make white space" for comparison overplotting (also can use z-limits)

Numerical - select numerical limits

– as with zoom~image_numerical, the limits are preserved



The image shows a vertical color bar with a gradient from black at the bottom to white at the top. To the right of the bar are three sets of numerical indicators. The top set is for the Z-axis, with a box at the top containing '100.' and a box at the bottom containing '0.000'. The middle set is for the Y-axis, with a box containing '5.70' and a box containing '0.000'. The bottom set is for the X-axis, with a box containing '5.70' and a box containing '0.000'. Each set of boxes is preceded by a small icon: a vertical line for Z, a horizontal line for Y, and a cross for X.

Z-scale color bar

These boxes display the current minimum and maximum values

X: X-axis of images or spectra (*)

Y: Y-axis of images or spectra (*)

Z: Z-scale of images are indicated at the bottom and top of the color bar

* The scale adjustments work in spectra~ overplot modes, as well as single plot modes, and when *Zoom~thumbnails* is used.

* **The (x,y,z)-limits for images and (x,y)-limits for spectra can be changed by entering values in the appropriate limit indicator box.**

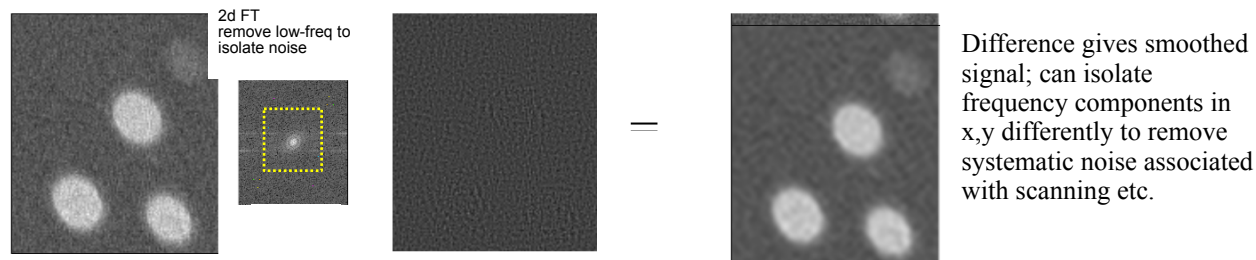
FILTER (for images and spectra)

5 types of smoothing with selectable parameters. **Clean (FT image filter)** works only with images. The other four apply to BOTH images and spectra.

Smooth – Boxcar average over n-points (right to edge of images) in odd

Clean (FT image filter) - 2d-FT filter. The FT is displayed on a 1:1 pixel format. Use the rubber-band style cursor (click, drag mouse, click a second time) to define the data in the complementary frequency domain to delete. The reverse transform of all data but the rejected data is displayed in buffer 9. The centre of the FT image corresponds to 0 (dc), while positions farther from the centre correspond to higher frequency. Periodic (moiré) noise associated with aliasing (beating) of a systematic noise signal with stxm sampling can be cleanly removed by deleting strong (typically linear) signals in the FT.

The **Clean (2d-FT Image filter)** can be used with subtraction of the filtered result to perform high-pass or selective band-pass filtering. For example -



Convolve - convolutes

with user-definable digital filter (definable frequency response)

Deglitch - -graphics-driven deglitch routine. Cursors identify lower and upper bounds of data.

Despike – local filter to remove point (spike) artifacts. User selects width of a median filter to identify the spikes. (NB the “**Despike stack**” routine in **Jacobsen’s stack_analyze** works better)

Median – n-point Savitsky-Golay averaging, except at edges

Lee Filter – Lee filtering smooths image noise by generating statistics in a local neighborhood and comparing the result to the expected values

Smooth
Clean (FT image filter)
Convolve
Deglitch
Despike
Median
Lee Filter

IMAGES (processing of images)

Add

- **Append** - append 2 images, matched on basis of (x,y) scales - images can be **tessellated** using append.
- **Buffer** – weighted addition of two buffers
(use a negative weight to subtract buffers)
- **Constant** - Add a constant to each pixel of an image
(use a negative constant for subtraction)

Average pixels - compute average intensity in a user-defined region

whole image – all pixels - full image

whole image - ignore zeros - same, but does not include zeros

region- all pixels - select pixel region of interest; report average Z-value with statistics

region- ignore zeros - as for all pixels, but does not include zero values. This is useful if an image has been multiplied by a [masked image](#) and the areas of interest are not contiguous. Use to count pixels and thereby determine areas of selected regions of an image.

bin – reduce image size (& improve statistics) by binning (2x2), (3x3), etc pixels to 1 pixel

Calibrate XY - calibrate X,Y scales of images

- **1-point** – shifts current image to make selected point have user-defined (x,y)
- **2 points** - allows linear stretching as well as shifting

NB: The calibration routines are similar to those used in Stacks~Image alignment. They can be used to manually process the first and last images of a sequence in order to ensure that the x,y limits selected will be present in all files.

*Note that once the parameters have been entered to calibrate ONE image, these are saved, so it is easy to apply the same calibration to a number of images, as a manual alignment process. For example, if a few-image stack is measured one energy at a time, **Calibrate XY** is an easy way to generate roughly aligned images. One can then write out the xy-calibrated images, then make a stack list (*.sl) file using a text editor, like NotePad, then use stacks~Analyse~stack list input to assemble the set of xy-calibrated images into a stack, which can be aligned further (if needed) using the Jacobsen or Zimba stack align routines.*

Example of a stack list file: [1st line is path to data; successive lines are the file names]

```
Y:\aXis-Pass-Test-data\05-STACKS\00-ANALYZE\01_STACK_LIST_INPUT\
12_055.axb
12_056.axb
12_057.axb
12_058.axb
12_059.axb
```

Add	>
Average pixels	>
bin	
Calibrate XY	>
Clip signal	>
Change	>
Convert to_OD	
Delete region	
Distort XY scale	
Fix rollover	>
FRC - resolution	
Gain	>
generate mask	>
histogram	
Modify one point	
Modify X,Y axes	
Multiply buffers	
Particle analyze	
Power	>
Profiles	>
Ratio to	
Remove zeros	
Replace line	>
Resize	
Set XY scale	
Transform	>
Warp	

Images (continued)

Change

dwell – change dwell per pixel (ms)

energy – change photon energy (eV) or equivalent parameter

header – modify header (this is what will be saved to disk)

mesh - redefine numbers of pixels (images) or points (spectra). This is useful to match pixelation of different images. Many routines in aXis2000 can modify pixel count by 1 or 2 pixels. *Change~mesh* can be used to make(x,y) pixelation of a set of images the same

x_label – change X- label (scale bar, energy, dwell)

y_label – change Y-label (polarization,

dwell
energy
header
mesh
x_label
y_label

Clip signal

replace all values outside user-selected limits with a fixed value (default is the average of the in-bounds data) . select the lower & upper limits by

Histogram – use cursors on a plot of a histogram of z-values

Numerical – use text input

histogram
numerical

Convert_to_OD - compute [OD](#) (optical density) representation of a transmission image with user defined I₀ value. The default is the maximum value pixel in the image (typically this is too large – should use *image~average pixels~region- all pixels* in a hole area of the image to get a more precise OD)

Delete region – use box cursor to define a region to replace with user selected value.
(this is useful to remove glitches or to select part of a masked region).

Distort XY scale – distort (x,y) scaling. Pixels interpolated to square in new co-ordinates

Fix rollover - for some image files (e.g. ALS PEEM) it is possible to have signed or unsigned formats which leads to 'roll-over' for pixels with above 16 K counts (e.g. if an unsigned integer data set is read using a signed integer). This replaces all pixels with a -ve value with 65,535 plus that value. Similarly, in phase outputs from ptychography reconstructions, many reconstruction programs automatically 'fold back' phase values such they lie between $-\pi$ and $+\pi$

2pi – replace points after increase of 2pi by previous value -3.14159
- replace points after decrease of 2pi by previous value +3.14159

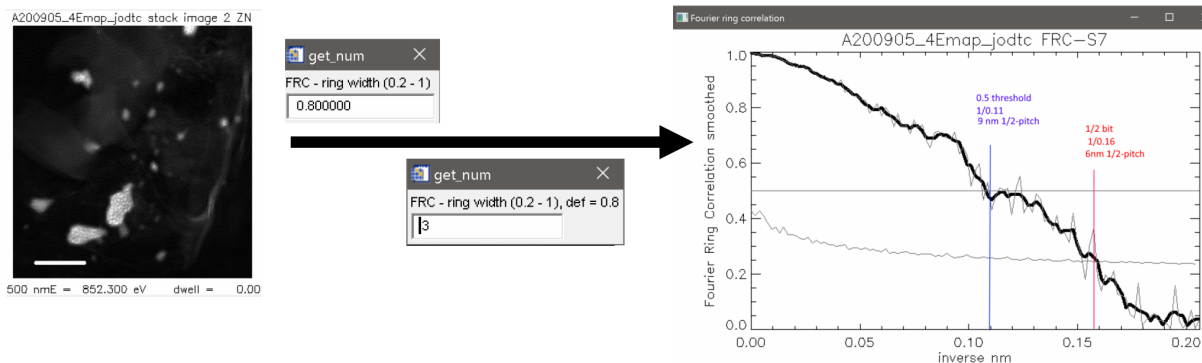
2pi
2**16
user_defined

216** replaces all pixels with a -ve value with 65,535 plus that value

user_defined – user specifies the 'rollover' threshold criteria and the value to increment (if pixel value goes -ve across threshold) or decrement (if value goes +ve across threshold)

Images (continued)

FRC – resolution – apply Fourier Ring Correlation algorithm to a square image to estimate spatial resolution. FRC curve crossing 0.5 and the 1/2-bit line are used to estimate the 1/2- pitch spatial resolution. For the equivalent full width at half maximum (FWHM) multiply by 2.



Gain - Multiply - multiply the Z-values by user-supplied constant

Divide - divide the Z-values by user-supplied constant

Multiply
Divide

generate mask – generates a **0/1 map** (1 for selected pixels; 0 if not-selected)

writes a ROI file

these maps be used in conjunction with **Multiply buffers** to select portions of images

optional write of a region_of_interest (ROI) file to allow selection in **stack_analyze**

threshold
manual

threshold - histogram

allows adjustment of lower and upper limits while seeing selected regions change dynamically (red overlay)

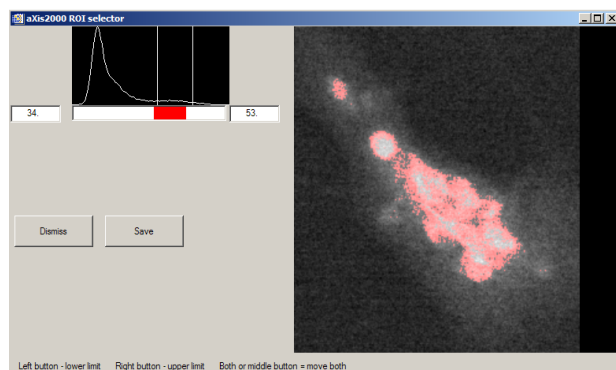
threshold –minimum user inputs the lower limit only (NB must be >0)

histogram
minimum

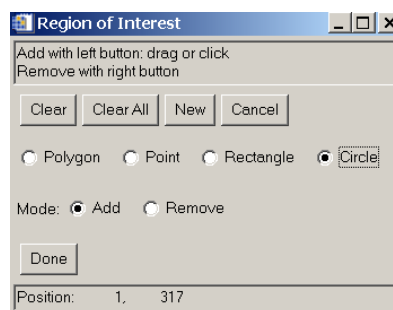
manual – use region definition tool to select a particular region for stack area selection.

NB this ROI selector is also used in e.g. *images~delete region* and *stacks~analyze~Zimba*

threshold region selector



manual region selector



Images *(continued)*

histogram – compute the histogram of all the pixels in an image

Modify one point – allows modification of a pixel (NB use **Delete region** for area modification)
Left click to add, right click to delete; in each case prompt is a local average

Modify x,y axes - applies a scale factor to the numerical axes of an image
(useful if there is a known distortion of a scanning system)

Multiply buffers – take product of the current and a second, user selected buffer

Particle analyze - when applied to a [masked image](#) (0/1 pixel values only) it analyzes the areas of contiguous regions and reports a distribution of diameters, assuming circular areas.

Power

- **exp(Z)** – exponential of z
- **ln(Z)** - natural log of z-values
(use to convert Transmittance to Absorbance)
- **10^(Z)** – raise z-values to 10
- **log10(Z)** - base-10 log of z-values

exp(Z)
ln(Z)
10^(Z)
log10(Z)

Profiles - generate and save [intensity profiles](#) from images

Linear – intensity along line defined by two user selected points

Radial – intensity as a function of angle in a circular region. The user defines a diameter which is then rotated about its center. Optionally, the resulting ‘unfolded’ radial distribution can be symmetrized by auto-seeking a threshold level and aligning at that common level. (This routine was written to explore [radial distributions in particles](#). It is also useful for analysis of azimuthal orientation effects probed by linear dichroism.)

Ratio to - computes ratio of 2 buffers with optional scaling. Images are matched by (x,y) scales

Remove zeros – replaces zero value pixels with a local average. (despeckles images with dropouts)

Replace line

Auto automatically identifies ‘bad lines’ (ones with pixels that deviate beyond expected statistical fluctuation (3 sigma) and replaces those lines with adjacent line(s)

auto
manual

Manual - use cursor to identify horizontal lines to remove. The selected line will be replaced by the average of 2 adjacent lines (default) or by any selected line, as the user chooses. The line suggested is the (n+1) line (the one above the selected line). Cursor line value is indicated during replace line selection. This is useful for images which are acquired line-at-a-time which can fail on the triggering of a line resulting in a line with only ‘0’ values. Numerical selection of the line to replace allows access to un-displayed lines in large images. Use the [pixel indicator](#) to determine the line number

X	263.05	9
Y	701.08	20
Z	12311.	

Images (*continued*)

Resize - set size of image in terms of # of pixels explicitly (this is needed to correct rounding errors in re-pixelation steps (e.g. shifts, truncates etc) whenever 2 or more images have to have identical numbers of (x,y) pixels for subsequent processing (same as *utilities~change mesh*)

Set XY scale - calibrate distance scale by defining distance between two user-selected points

Transform

Flip
Mirror
Rotate

Flip – invert image vertically through a horizontal centre line

Mirror – invert image horizontally through a vertical centre line

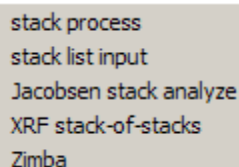
Rotate - rotate image about a user defined point, by user defined angle. This can be used in conjunction with *Images~Distort X,Y scale* to remove image distortion by symmetrizing an object of known shape (e.g. circular particles).

Warp –uses multiple matching point “morphing” capability of IDL. Intended to handle cases where a 2-point or 1-point manual shift will not compensate for changes in the sample shape in a stack (the ‘warping’ usually occurs because of radiation damage).

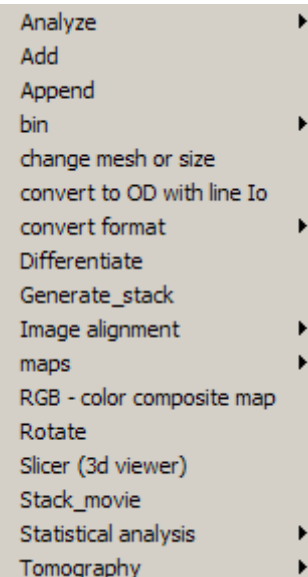
STACKS - manipulation of image sequences.

Note, the ability to simultaneously manipulate multi-dimensional data sets (3,4,5 & more) is one of the strengths of aXis2000, combining aspects of both image and spectral manipulation.

Analyze



- stack process
- stack list input
- Jacobsen stack analyze
- XRF stack-of-stacks
- Zimba



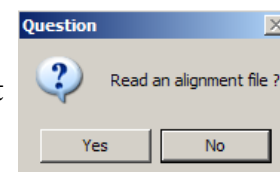
- Analyze
- Add
- Append
- bin
- change mesh or size
- convert to OD with line Io
- convert format
- Differentiate
- Generate_stack
- Image alignment
- maps
- RGB - color composite map
- Rotate
- Slicer (3d viewer)
- Stack_movie
- Statistical analysis
- Tomography

Stack process

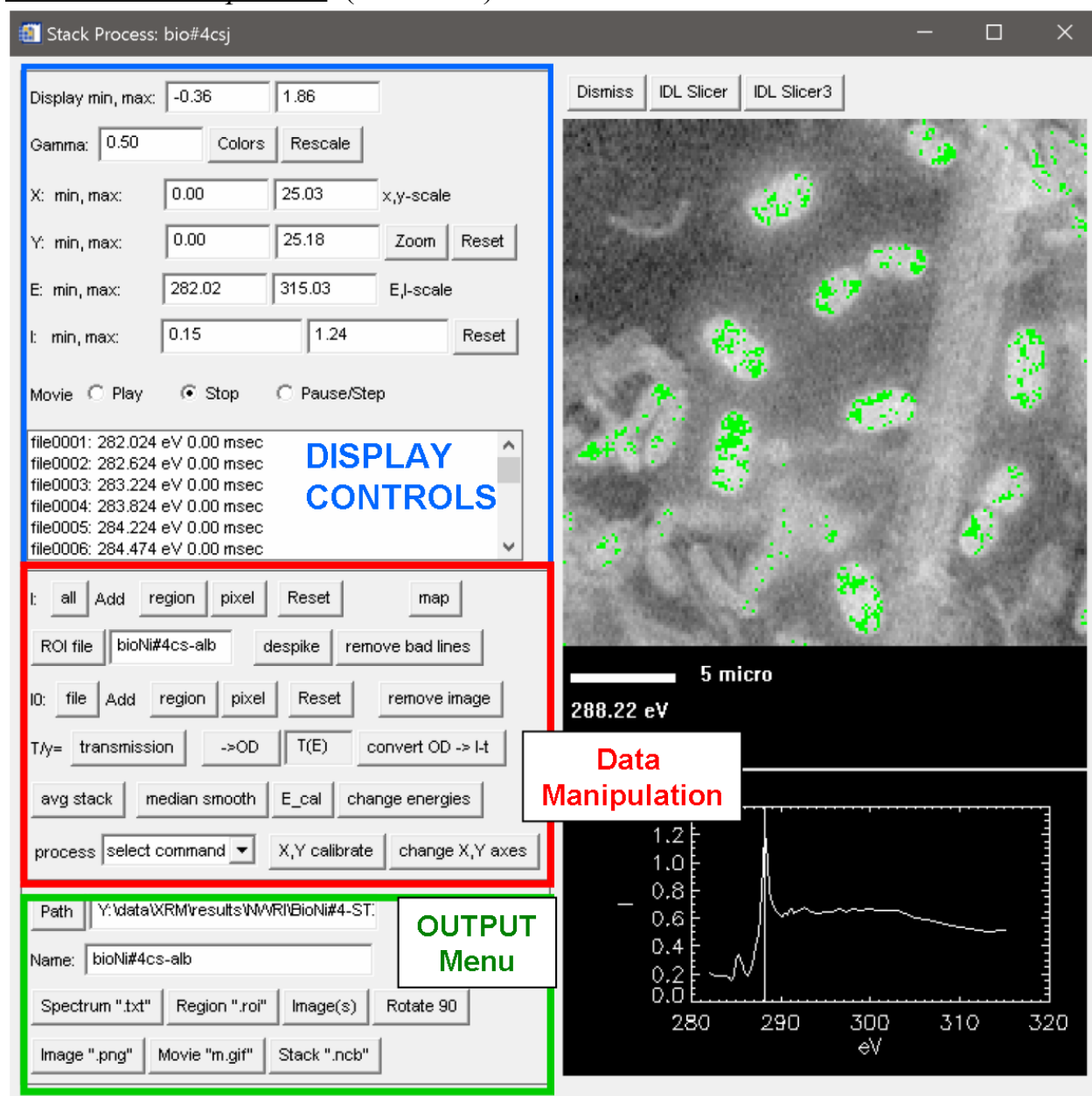
- loads an AXIS binary stack format file (*.ncb) into the **stack_process** widget. Note that it is convenient to store image sequence data in this binary representation since it is much more compact (by three times or more) than original data formats. Since it is possible to extract images from the binary file and since the IDL cross platform binary formatting is used, no information is lost.

The user is prompted for an alignment file (to incorporate alignment shifts into data) and a **widget size scale factor**, before the following widget appears. The *.aln files are written by the Zimba_align or stack_align (early Jacobsen) procedures

The default **widget size scale factor** will make the stack_process widget fill ½ of the computer screen. For very large stacks, one can use a fractional **widget size scale factor**. This will display and allow visualization but many of the data modification actions of stack_process do not work with fractional **widget size scale factors**.



STACKS~stack_process (continued)



Notes

1. click on spectrum to select image at that energy (or click in the image list)
2. process 'select command' pull down menu contains many full-stack actions:

select command

+/- constant
+/- spectrum
+/- image
* constant
* spectrum
* image
/ constant
/ spectrum
/ image

STACKS~stack process (continued)

DISPLAY CONTROLS

Display min, max – Z-axis limits

Gamma – gamma for Z-axis

Colors – select from palette of standard IDL color tables

Rescale – [ON] min/max each image. [OFF] – use the display min, max settings

X: min, max – use to set the x-scale of image (for precise region selection)

Y: min, max – use to set the y-scale of image

Zoom – graphical expansion of image **Reset** restore full stack (x,y)

E: min, max – use to set the energy range of the spectral display

I: min, max – use to set the intensity range of the spectral display

Reset – reset (E, I) scales to full scale values

Movie – play – play the movie

- **stop** – stop playing movie
- **pause/step** – pause, or advance one frame

Image parameter list –select image by clicking or using arrow keys to scroll up/down

DATA MANIPULATION CONTROLS

I: all - display average spectrum of all pixels (treated as I in Beer's Law or TEY)

Add **region** - display average spectrum of an I region selected by polygon draw selector

pixel - display average spectrum of a selected pixel

Reset – reset selection of I pixels

map – use 2 (A-B) or 3 (A-(B+C)/2) selected images to take make a chemical map

ROI file – click to browse & select a *.roi (region of interest) file

despike - replaces zero value pixels with a local average.

remove bad lines – replace lines ~ auto applied to all images in a stack

Io: file - browse to select spectrum as Io (treated as Io in Beer's Law or TEY)

Add **region** - display average spectrum of an I region selected by polygon draw selector

pixel - display average spectrum of a selected pixel as Io

Reset – reset selection of Io pixels

remove image – remove currently displayed image from the stack

T/y = switch between transmission and yield (electron, XRF etc) modes

->**OD** – convert Z-values from transmission to optical density using currently defined Io

T(E) toggle between absorption ($-\ln(I/I_o)$) and YIELD type TEY (I/I_o) normalization

(the current status is displayed between ->OD and TEY boxes)

convert OD → I-t – convert absorbance to transmittance using user-supplied Io

avg. stack – compute average of all images within the DISPLAYED E-min/ E-max range
& store as a file and in a selected aXis2000 buffer

median smooth – apply 3-point median smooth to all images

E_cal – calibrate the energy (linear shift by user selected amount)

change energies – exchange current energies of stack for those in a selected file

process **select command** – modify stack (see next page for description of commands)

X,Y calibrate – define width of stack area (aspect ratio is preserved)

change X.Y axes – replace present (x,y) values with those from an external file

STACKS~stack process (continued)

process **select command** :

- +/- **constant** – subtract a constant from z-values of each image
- +/- **spectrum** – subtract a spectrum (*.txt) from spectrum at each pixel
- +/- **image** – subtract image from file (*.axb) from each image in the stack
- * **constant** – multiply each image by a constant
- * **spectrum** – multiply stack by a spectrum (*.txt)
- * **image** – multiply each image in stack by an image (*.axb) (use for masking)
- / **constant** – divide each image by a constant
- / **spectrum** – divide stack by a spectrum (*.txt)
- / **image** – divide stack by an image (*.axb) (use for masking)

select command

+/- constant
+/- spectrum
+/- image
* constant
* spectrum
* image
/ constant
/ spectrum
/ image

OUTPUT MENU

Path – defines the path for input / output files (default is folder that contained the stack)

Name – filename to be used for output

Spectrum “.txt” – write displayed spectrum

Region “.roi” – write currently selected pixel set to file (multiple regions are supported)

Image(s) – write displayed image

or all the images to a variety of formats

Format (0=*.nc, 1 = *.axb, 2 = *.bmp, 3 = *.tif(long), 4 = *.tif(float)), 5 = text
1

Rotate 90 – write out stack after rotating the stack clockwise or counterclockwise by 90°

Image “.png” – write current display (image and spectrum) to a *.png format file

Movie “.gif” – write images or {images & spectra} as a movie

The stack_analyze routines generate a multi-gif file which is 'clunky' and the *.mgif file written does not play due to licensing issues. The **AVIMaker** from Platypus software (<http://www.c-point.com/>) can be used to make *.avi files from the set of *.gif files written by the Movie command. Alternatively a *.gif movie can be created using the Easy Gif Animator (<https://www.easygifanimator.net/>). The individual gif images are required as input so answer 'N' when asked, “delete all *.gif files?”

ALSO – while the routine is SUPPOSED to store the images in the default folder (the one containing the stack), it often ends up in a different folder, typically the source code folder (C:\aXis2000), and sometimes the (n-1) active folder. Good luck hunting for the *.gif files !!!

Stack “.ncb” – write the stack data within the displayed E-range as a *.ncb stack. *Thus to extract a subset of the energies of a stack, modify the E-range by setting E-min/E-max in the ‘Display Menu’.* The spatial region stored is selected by defining the row / column pixel values (default is all area of the stack). The user is asked to define the lower and upper row and column indices for the saved data, thus allowing removal of the edges of a stack which might be distorted by the alignment procedure. The *.ncb binary file generated is needed for the **stack-fit** and **stack-SVD** routines. The save stack “.ncb” routine saves whatever is currently displayed in stack process. In order to use curve fitting routines with reference spectra it is necessary to normalize to I₀ and convert to OD scale (transmission measurements) or divide by an appropriate reference for yield data and then save the stack.

STACKS~stack_process (continued)

COMMANDS AT THE TOP RIGHT OF THE **STACK PROCESS** WIDGET

Dismiss – exit `stack_process` widget

IDL slicer – start widget to manipulate 3d display of stack (early version if the slicer widget)

IDL slicer3 – advanced 3d manipulation of stack using latest IDL Slicer widget

Stacks~Analyse (continued)

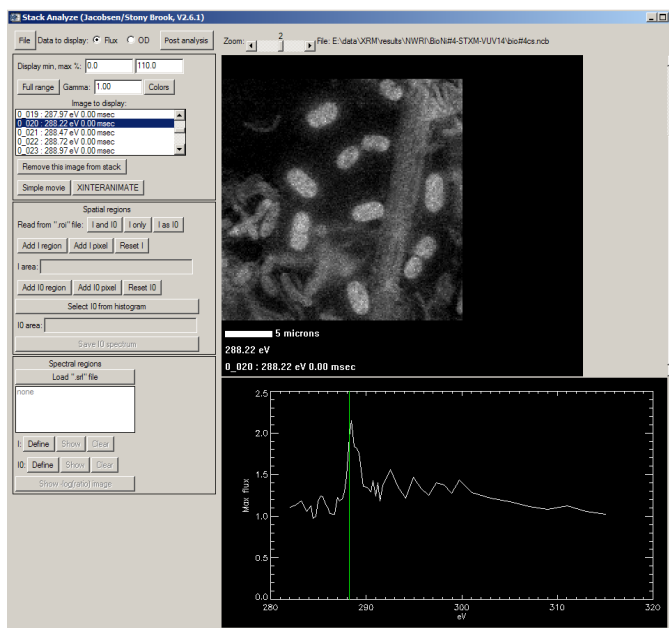
stack list input

- generate a stack from a `stack_list` file (*.sl) and (optionally) alignment (*.aln) files. If you cancel after selecting the first file, then `stack_build_list` is started. If you select a `stack_list` and then cancel after selecting the second file, then `stack_align` is started (note this has different alignment properties than either `stack_analyze` or `Zimba` aligners, but is generally inferior to either of those routines). Once these choices are made the **stack_process** widget (described above) is started. This command is used to assemble a stack from individual images or to re-assemble a stack, which has been written out as separate files, and has, for example, some modified images. Remember to write out the re-assembled stack !!

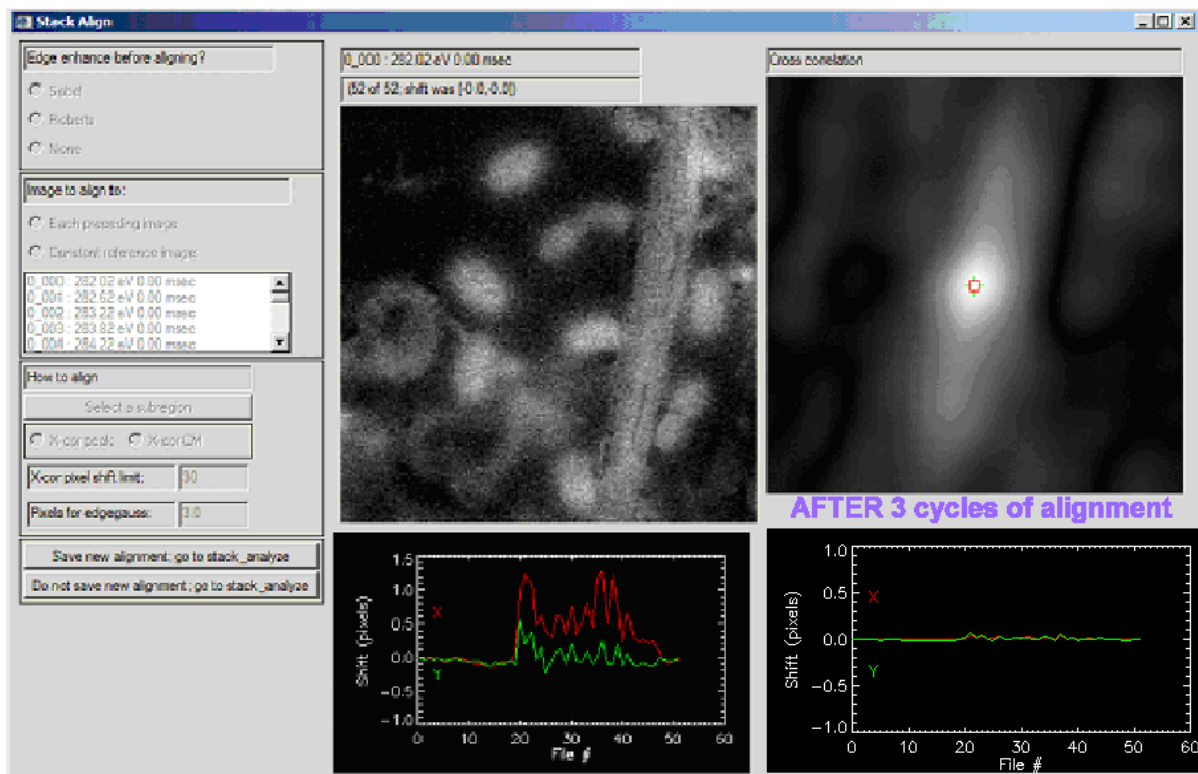
stack process
stack list input
Jacobsen stack analyze
XRF stack-of-stacks
Zimba

Jacobsen stack analyze – starts the Jacobsen `stack_analyze` widget, which is a **complete spectromicroscopy data processing package**. The version accessed by aXis2000 is from Sep 2008. More recent versions may exist. See the documentation for that widget, from Chris Jacobsen's web site. The capabilities of this package are extraordinary. The alignment routine is exceptional. Apply it many times until the (x,y) deviations are less than 0.1 pixel. After alignment, trim (Save autoclipped stack '.stk' file) and then Save axis '.ncb' file.

Menu under 'File'



STACKS Analyze ~ Jacobsen stack analyse (continued)



XRF stack-of-stacks – allows extraction of one multi-incident-photon-energy stack from an array of XRF stacks recorded at a set of incident photon energies. (Ultimately to be evolved into a true 4D viewer and processor widget – *anyone want to help* ?)

Zimba – Carl Zimba's version of the stack analyze code. It has a very nice alignment which is able to align stacks for which the Jacobsen alignment routine fails. For unknown reasons, when called from aXis2000 the Zimba routines often lock up if complex combinations of commands are used. In such cases the full functionality of the Zimba routines can be accessed by running *zstack.pro* outside of aXis2000 (if you have an IDL license) in which case you can access all the many features for display and storage, including saving a movie of a stack with display of the spectra from multiple regions.

Some features of the Zimba stack analyze package:

- Reliable alignment (which differs from the routine in Jacobsen's stack analyze).
- Many different formats for I/O data are supported (*.csv, *.txt, *.xas)
- Simultaneous display of original and aligned data
- Excellent manual alignment (ztune)
- Multiple spectral regions defined
- Provision to write full, aligned stack to a single file (*.ncb, *.stk)
- Provision to write image sequences as MPEGs, or a set of image file

STACKS Analyze ~Zimba (continued)

Note: The Zimba *.sl and *.aln file formats differ from those for the Jacobsen set. It is not generally possible to 'mix-and-match' the two sets without using a text editor for conversion. In particular, it is essential to delete all letter and non-numerical symbols from the lines listing the shifts in the *.aln file written by Zimba, before using it in the read-in procedure for stack_analyze. The ZSTACK codes can be run independently from AXIS.

Please see separate documentation for ZSTACK supplied by Carl Zimba included at the end of this manual.

Add – add two stacks (useful to take differences to remove dominant contributions to better display minority signals by using negative weighting)

Append – combined 2 stacks (must have the same pixel dimensions !) and order the images according to the photon energy (or equivalent variable)

bin - bins (2-2x2 to 1 pixel, 3 = 3,3 to 1 pixel, etc) image data for better statistics. The first character of the original file name is converted to a b to indicate the processing.

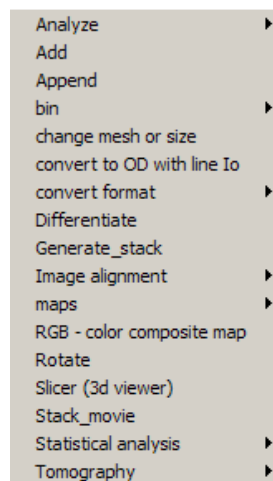
- **energy** - bin the data along the energy axis

- **NSLS - 1** – bin 1 user-selected *.nc file

- **file** – bin all files listed in the supplied file (typically *.sl files).

- **stack *.ncb** – bin all images in a stack

lox – bin all images in a Lox stack (PEEM from CLS)



1
file

*.ncb
lox

change mesh or size – adjust pixilation or the real-space dimensions

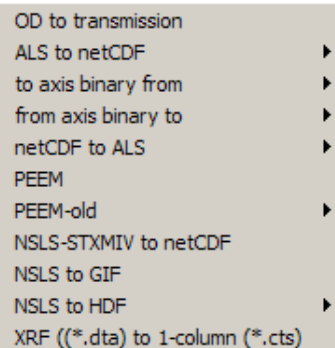
convert to OD with line Io – generate OD version of a transmission stack for the case where there is a set of columns all in the Io region, which can then be used to generate an Io on a line-by-line basis (normally a single Io value for each stack image is used in normalization). This routine is useful if the stack images are acquired so slowly that there is significant change in e.g. the ring current during acquisition of each image.

STACKS (continued)

convert format

For most conversions, the user selects the first file then the last file in a sequence; intervening files are converted assuming a standard file name convention (e.g. last 3 positions before '.' are an index number).

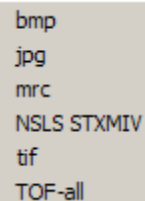
- **OD to transmission** – convert to transmission using “white” Io (1)
- **ALS to netCDF** – converts (old) ALS images to NSLS [netCDF](#) files, with E-scale shifts and optional binning (adds squares of 2x2 or 3x3 etc pixels). Generates *.sl file for input to Stack_analyze
 - **1** converts one file selected by pickfile
 - **many** converts many files selected by (first, last) pickfile routine
 - **file** converts files listed in the user-defined file (e.g. *.lst from ALS)
- **to axis binary from**
 - **bmp**
 - **jpg**
 - **mrc** (format used for tomography in, e.g. IMOD)
 - **NSLS STXMIV**
 - **TOF-all** (time-of-flight PEPICO)
- **from axis binary to**
 - **mrc**
 - **mpg**
- **netCDF to ALS** - converts NSLS [netCDF](#) format files to ALS format files
 - **1** converts one file selected by pickfile
 - **file** converts files listed in the user-defined file (e.g. *.lst from ALS)



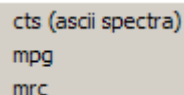
OD to transmission
ALS to netCDF
to axis binary from
from axis binary to
netCDF to ALS
PEEM
PEEM-old
NSLS-STXMIV to netCDF
NSLS to GIF
NSLS to HDF
XRF ((*,dta) to 1-column (*.cts))



1
many
file



bmp
jpg
mrc
NSLS STXMIV
tif
TOF-all



cts (ascii spectra)
mpg
mrc

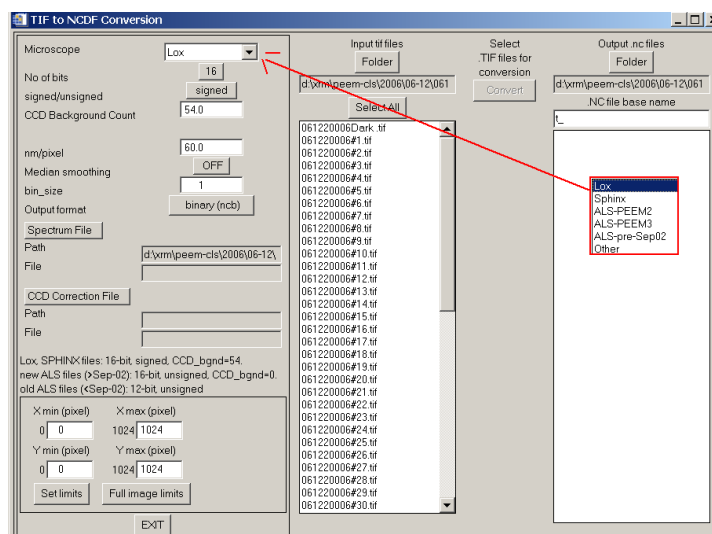


1
file

STACKS~convert format (continued)

PEEM - converts PEEM stacks written by various instruments (Lox, Sphinx, AALS-PEEM2, ALS-PEEM3, ALS-pre-Sep02 (peem2 old format), Other)

Recommended for ALL types of PEEM data



The output is either a binary stack (*.ncb) (read with *stack_process*), or as a set of {*.nc} images (read in with *Stacks~Analyze~stack list* input (or *Zimba*). The default conditions for SPHINX, ALS-PEEM2 [new (>Sep-02) & old (<Sep-02)], ALS PEEM3 and Lox are indicated in the left centre of the widget.

The upper left part of the widget sets the conversion parameters

nm/pixel – sets spatial scale

No of bits – 12 or 16 bit

Signed/unsigned

Median smoothing – Yes/ No

Bin - reduce size and improve statistics by 2x2 => 1; 3x3 => 1 etc

Lox - Yes / No

Output format {*.nc} set with *.sl stack list, or *.ncb file

CCD Background count – signal in absence of X-rays

Input files **Folder** – select folder with data to convert to stack (lists *.tif files)

Select all – highlights all files (alternatively select those

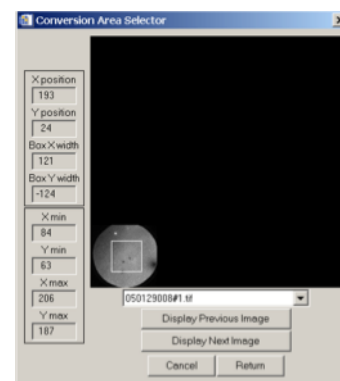
you wish to convert

Select CCD Background file – for patterned dark signals

Select spectrum file – get energies of stack from spectrum generated during stack acquisition

Get Limits – identify pixel range to convert

Convert – this button will only become active after an energy spectrum is identified and the files to be converted have been selected. After pushing this button the files are converted to *.nc files, either temporary, or permanent. If Output format is set to *.ncb, only the binary stack is written to disk.



STACKS~Convert format (continued)

PEEM-old (ignore unless desperate!)

- ALS-PEEM

to *.ncb – convert to binary stack file, without writing individual netCDF files

PEEM-old	ALS-PEEM	to *.ncb
NSLS-STXMIV to netCDF	SPHINX	to netCDF

to netCDF – convert to set of *.nc (netCDF) files and a stack list (.sl) file

This activates the ALS PEEM read-in widget (see [Read~images~ALS PEEM](#)) and the user selects the first and last files, all conversion options, then executes the conversion.

Note: this widget does not work in the IDL VM version

- SPHINX - convert 16-bit TIF files from SPHINX LabView program (Elmitec at SRC)

to *.ncb – convert to binary stack file, without writing individual netCDF files

to netCDF – convert to set of *.nc (netCDF) files and a stack list (.sl) file

This activates the ALS PEEM read-in widget (see [Read~images~ALS PEEM](#)) and the user selects the first and last files, all conversion options, then executes the conversion.

Note: this widget does not work in the IDL VM version

NSLS STXMIV to *.ncb – converts nsls STXMIV image sequence directly to a binary stack file (all images in one file). NB One can use Zimba or Jacobsen align procedures on *.ncb files.

NSLS-STXMIV to netCDF - convert NSLS stxmIV format files to netCDF format files

NSLS to GIF – converts one *.nc file to a GIF image

NSLS to HDF - converts *.nc files to HDF format data files

1 converts one file selected by pickfile

many - converts a sequence of files with first, last identified by pickfile

OD to transmission	
ALS to netCDF	▶
to axis binary from	▶
from axis binary to	▶
netCDF to ALS	▶
PEEM	
PEEM-old	▶
NSLS-STXMIV to netCDF	
NSLS to GIF	
NSLS to HDF	▶
XRF ((*.dta) to 1-column (*.cts))	

XRF ((*.dta) to 1-column (*.cts))

Converts XRF spectra from Oxford system to readable values

Differentiate – compute the derivative of the spectra at each pixel in a stack

Expand - add additional pixels on left/right and top/bottom to a selected stack.

Fix rollover – as for Images, but applied to all images of a stack. for some image files (e.g. ALS PEEM) it is possible to have signed or unsigned formats which leads to 'roll-over' for pixels with above 16 K counts (e.g. if an unsigned integer data set is read using a signed integer). This replaces all pixels with a -ve value with 65,535 plus that value. Similarly, in phase outputs from ptychography reconstructions, many reconstruction programs automatically 'fold back' phase values such they lie between $-\pi$ and $+\pi$

STACKS *continued*

Generate_stack - a stack is computed and saved from an image and a spectrum, with the image and spectrum selected by the user from aXis2000 buffers.

Y_illumination correct ratio the first selected stack by a second stack (which is the illumination, which can be generated by heavy smoothing all the images in the first stack)

Image alignment - tools to manually align a series of sequential files (either ALS or NSLS format; read in after defining first and last of the sequence). *Note this always works whereas the manual align in ZSTACK is somewhat 'tempermental'*. There is a nice manual alignment tool on *Stacks~Analyze~Jacobsen stack analyse*

shift - 1-point calibration of image (x,y-scale).

stretch/ shift - 2-point calibration of image (x,y-scale)

Analyze	>
Add	
Append	
bin	>
change mesh or size	
convert to OD with line lo	
convert format	>
Differentiate	
Expand	
Fix rollover	>
Generate_stack	
y_illumination correct	
Image alignment	>
maps	>
RGB - color composite map	
Ratio to another stack	
Rotate	
Slicer (3d viewer)	
Stack_movie	
Statistical analysis	>
Tomography	>

After shifting or stretch/ shifthing the (x,y) scales for image alignment, this routine then:

1. grids to a user-defined pixel size (should be similar or smaller than the recorded pixel size to avoid loss of information)
2. truncates to a fixed user-defined [xmin, ymin, xmax, ymax].
3. bins to a user defined factor (1=no binning) to trade off S:N versus spatial resolution
4. writes a netCDF file. The names are FORCED to be the same as the input but with the 1st letter of the original filename converted to **s** (shift) or **a** (shift/stretch).
5. generates *.sl file of filenames (read using *Stacks~Analyze~stack list input*). In addition it stores 2 (**s**) or 4 (**a**) files (in buffers 5,6 and 8,9) which are the shifts in real-space units (microns) **The parameters defined in the first pass are saved and applied automatically to all subsequent files.** The X,Y shifts and stretch terms for each file are accumulated in buffers 7 (stretch) 5,6 (point 1,x,y) and 8,9 (point 2,x,y) for later use to explore how the microscope or the sample is changing.

Notes for image alignment:

- Clear buffers 5-9 (using the **Clear Buffer** above the thumbnails) before starting as they are used for results
- Check the extreme limits of a stack sequence to ensure the [xmin, ymin, xmax, ymax] values used to define the common part of all images are valid for all files.

Warp – Image alignment using a polynomial 2-d transformation to align successive images to the 4 or more fiducial points identified on the first image, and on each subsequent image. This can be useful when the sample distorts during a measurement due to radiation damage.

STACKS (continued)

maps

SVD - convert an image sequence (stack) to [component maps](#) using **singular value decomposition** (SVD) procedures. When accessed from this menu item, the **input must be an AXIS format binary file stack (*.ncb)**.

SVD
Stack fit
CGO curve fit
polarization fit

Typically the input stack consists of a set of images prepared on an optical density (OD) scale with careful alignment. The reference intensities at each energy of the stack are extracted from user identified reference spectra (*.txt, read from disk), which are interpolated / extrapolated as needed to match the stack energies. The user is prompted to either read the list of reference spectra from a *.par parameter file, or, after identifying number and filenames of the individual reference spectra, write the list to a *.par file. This is helpful in cases where the same set of reference spectra might be applied to a number of stacks.

The **output** is a set of **chemical component maps**, automatically written to files with names constructed from a root and the component names (it is wise to keep the component names SHORT to avoid excessively long file names). In addition the **residual** signals averaged over all energies is saved as an 'image'. Comparison of the magnitude and spatial distribution of the residual signal is a useful way to evaluate the validity of the analysis. Optionally a **stack of the residual images** (difference of data and fit at each energy) can be saved for later examination. If the input stack is in OD units, and the reference files are on absolute linear absorption scales (**OD1**, or optical density per nm thickness at the standard density of each component), the Z-values of the resulting component maps are in absolute thickness units (in nm, assuming the reference spectra are in units of nm⁻¹).

NB the SVD code can also be run from the IDL command line and has different features not implemented directly in aXis2000. See the code file for further details.

Stack fit – Performs a linear regression analysis (linear least squares fit) of the spectrum at each pixel to a sum of (1 to 8) user-defined model spectra and a constant.

NOTE: Relative to SVD maps, stack fit adds an additional component to the analysis, a 'CONSTANT' which is flat spectrally (same at all energies) but different at each pixel. This always leads to a better fit statistically (smaller residual, often by 2 orders of magnitude) may lead to systematic error in the result. Stack-fit rather than SVD should be used where there are offsets of reference spectra relative to the data, due to Io errors,

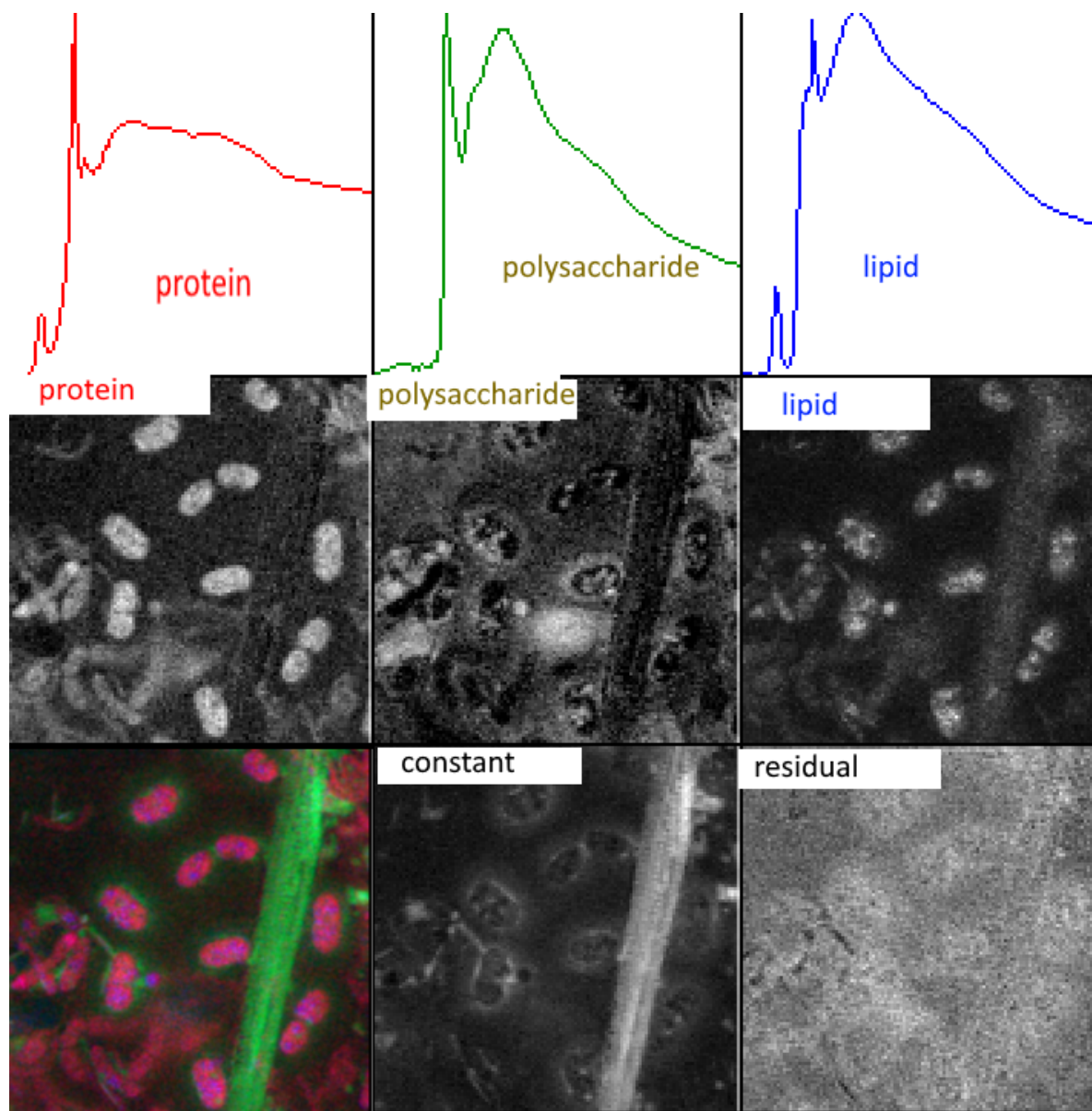
CGO curve fit - This uses a conjugate gradient optimization method (from Numerical recipes) to perform spectrum-by-spectrum curve fits to reference spectra. The user dialog is similar to those used in the SVD and stack fit routines. The fit of a single spectrum can be carried out using this method with the *spectra~curve fit* command.

In tests on low noise data sets with valid OD scales and with accurate spectral models, essentially identical component maps are generated by SVD maps, stack fit and CGO curve fit

STACKS~maps (continued)

EXAMPLE of using *stacks~maps~SF* to convert a C 1s stack to a set of component maps.

Data: wet biofilm (courtesy John Lawrence, National Water Research Institute, Saskatoon)



Reference spectra are quantitative (OD1) so z-scales on the **protein**, **polysaccharide** and **lipid** maps are nm thickness. **constant** represents signals from water, a silicate diatom, and CaCO₃ and K at the envelope of the cells. The color coded composite (buffer 7) combines the **protein**, **constant** and **lipid** component maps. The image area is 25 µm x 25 µm. (Lawrence et al, Applied Environmental Microbiology 69 (2003) 5543)

STACKS~maps (continued)

For **SVD**, **Stack fit** and **CGO-fit** analyses with less than 5 reference species, at completion the aXis2000 buffers contain:

Buffers **1-3(5)** – the reference spectra

Buffers **4-6(8)** – the component images - i.e. the spatial distribution of the component

Buffer **7** – the map of the linear term (for **Stack-fit** only)

Buffer **9** – the map of the chi square values of the fit at each pixel (~ fractional uncertainty)

If there are more than 4 components, the spectra are displayed, but then the component maps are displayed in sequence, starting in buffer #1/

For analyses with more than 5 reference species, the component maps are stored in the same buffer number (**1-8**) as the reference spectra and buffer 9 contains the residuals map.

After making the component maps it is often useful to explore the spatial correlations of components by combining any 3 of the component maps into a single **color-coded composition map**, by using the *Display~RGB composite* command or (if you are running a IDL licensed version) *stacks~RGB color composite map*

HINT: If you want to create a 1-color or 2-color composite, select a buffer that is empty or a spectrum for the colors you do not wish to include.

How to evaluate the significance of maps derived using SVD, Stack fit or CGO fit.

The user of *stacks~maps* must realize that the fitting code **will always give a result, but the result may not be valid**. It is important to evaluate the quality of the fit. aXis2000 provides a number of tools to do this.

* Examination of the **residual stack** can help to evaluate the validity of the fit. Spectra extracted from various regions of the residual stack should be only noise; if there is a missing chemical component one can sometimes obtain its spectrum in the poor fit regions of the residuals stack.

* One can apply *images~generate_mask* on the **residual map** (in buffer #9) to write out a *region_of_interest* (ROI) file corresponding to the poor fit regions. The ROI files can be used in *stack_process* to extract the spectrum of the poor fit region, which may be a 'missing component'.

* One can apply *images~generate_mask* on the **component maps** to generate the ROI of the pixels with large amounts of components of interest. After extracting the spectrum of those pixels using the ROI files in *stack_process* the **quality of the spectral fit** can be examined by applying *spectra~curve fit~{linear regression (=stack fit) , or CGO(=SVD)}*

* An alternate method, such as a multivariate statistical analysis (MSA) approach- e.g. PCA_GUI (*stacks~statistical analysis~PCA_GUI* (CJJ Dec 2005) or **Mantis** (<https://spectromicroscopy.com/>) can be explored, If the results of the forward fitting approach (maps~SVD or stackfit) and MSA methods are similar, greater confidence can be placed in

STACKS~maps (continued)

Polarization fit

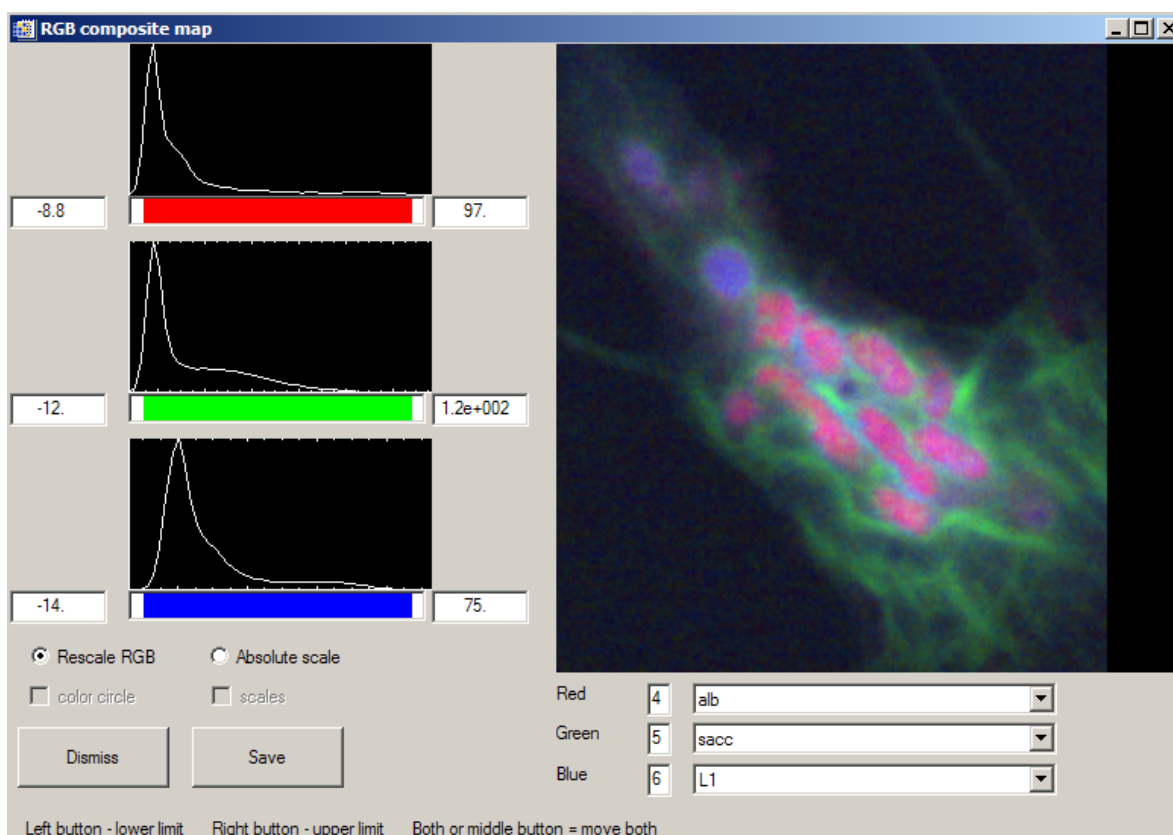
Fits a sequence of images where the control variable is the angle between the E-vector and the image. Such data can be generated by azimuthal angle scanning (e.g. at STXM532), or by rotating the E-vector with an EPU (at STM1102 and CLS-STXM). The polarization signal is fit to

$$I(\theta) = C + A * \cos^2(\theta - B)$$

where C is a non-angle dependent constant, A is the amplitude of the dichroic signal and B is the 'director' (angle of maximum intensity)

Note that the quality of the fit can be evaluated by selecting a region of interest, extracting the angle-dependent signal then using *spectra~curve fit~pol fit*

RGB Color composite map – uses a widget with lower / upper limit controls and selector of any of the aXis2000 buffers to visualize spatial arrangements of chemical components (e.g. as derived from curve fitting (stacks~maps) or multivariate statistical analyses (or simply, images at 3 different energies)



Note: The **RGB Color composite map** does not function properly when executed from the virtual machine (VM) version (tested Jan 2023, on IDL6.3 and IDL8.6). Use *Display~RGB composite*.

STACKS (continued)

Ratio to another stack – divide the stack by a user selected stack (*.ncb) (NB stacks can be generated from an image and spectrum using *stacks~Generate_stack*)

Rotate – rotate the whole stack by user selected angle. Boundaries are filled with the minimum over each image

Slicer (3d viewer) – IDL's 3d viewer with read in of a binary stack

Stack_movie calls Jacobsen *stack_movie* routine. Files defined by user-selectable **stack list file (*.sl)**.

If 'cancel' is selected, the *stack_build_list* routine is initiated.

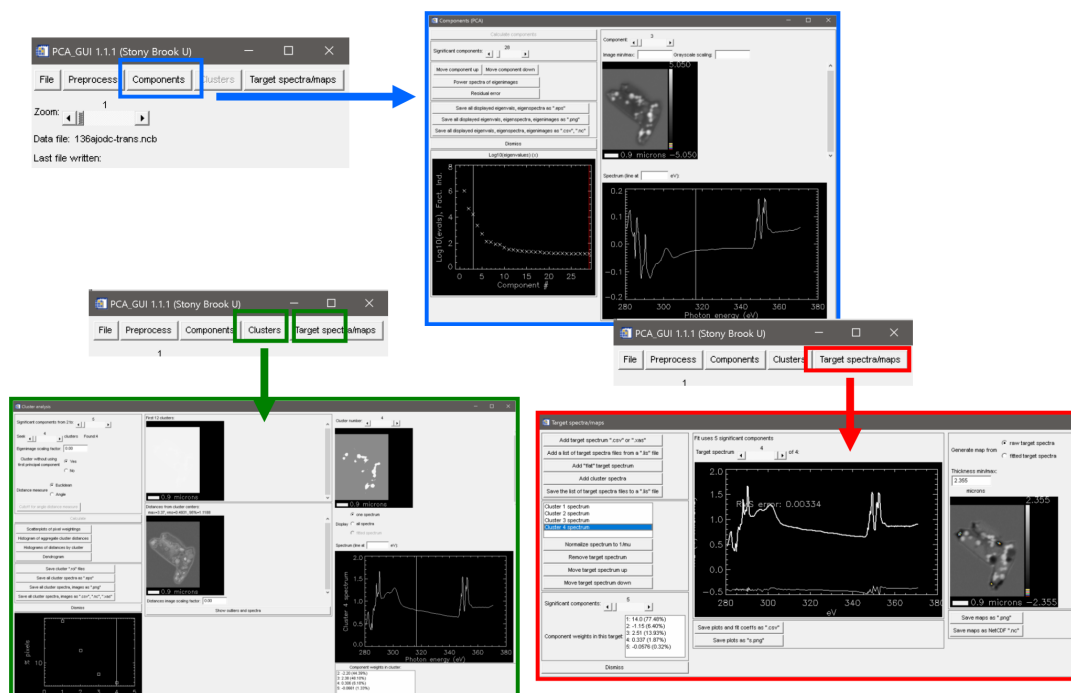
Statistical analysis

- **Principle components** - applies the IDL principle component analysis package (PCOMP.SAV) to derive the **eigenspectra** (power weighted representation of the data) from a set of OD images (binary stack in an *.ncb file). The routine implemented in aXis2000 provides the **eigenvalues** (fractional contribution of each eigenspectrum) and eigenspectra of the first 8 components. This is useful to obtain a sense of how many independent chemical species might be present in a give data set. The **eigenimages** are readily generated by using the eigenspectra as model spectra in a SVD map

- **PCA_GUI (CJJ Dec 2005)** – combination of principle component, cluster and target analysis allows for unsupervised analysis to extract a set of potential NEXAFS spectra, with extensive capabilities to evaluate and optimize. See the manual for this package for further details.

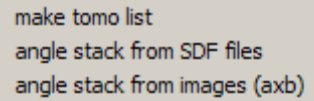
Analyze >
Add >
Append >
bin >
change mesh or size >
convert to OD with line lo >
convert format >
Differentiate >
Expand >
Fix rollover >
Generate_stack >
y_illumination correct >
Image alignment >
maps >
RGB - color composite map >
Ratio to another stack >
Rotate >
Slicer (3d viewer) >
Stack_movie >
Statistical analysis >
Tomography >

principle components
PCA_GUI (CJJ Dec 2005)



Stacks *(continued)*

Tomography Routines to analyse **4d** (x,y, E, θ) data.



make tomo list
angle stack from SDF files ▶
angle stack from images (axb)

Make tomo list – write a list of the folders containing tomo data.

This can then be edited to include the angles at which each stack was measured.

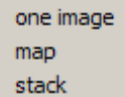
Angle stack from SDF files

reads a set of angle varying images to generate an (x,y, θ) stack

one image – select one image from a set of images (stack) at each angle

map – select a map from each folder

stack - read full 4-d (x,y,, E, θ) data and convert to a format suitable for read in to IMOD.



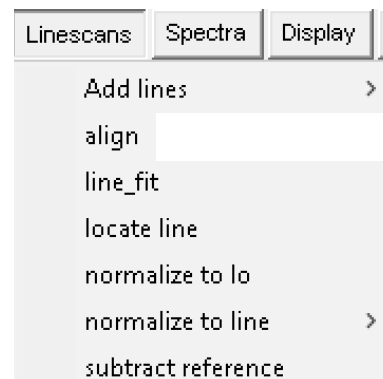
one image
map
stack

LINESCANS (processing of linescan spectra)

Processing of [linescan spectra](#). Linescan processing methods are also useful for a variety of image analysis tasks such as:

- obtain averaged **profiles** from an image along X or Y
- remove low frequency noise from images (average all horizontal, or all vertical, then ratio)

HINT: If the direction of interest in the image is not oriented along vertical or horizontal, use *Image~rotate* to rotate the direction of interest to horizontal or vertical

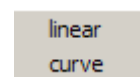


Add lines - sums all lines selected with cursor (lower / upper limits)

- **Horizontal** (for Linescans, sum spectra over a range of length along the line)
- **Vertical** (for Linescans, sum line profiles over a range of E)

align - 'tilts' data to user specified line

- **Linear** – straight line defined by 2 points
- **Curve** – curved line defined by multiple straight line sections



line_fit – apply SVD-based curve fit to a linescan (same method as in *stacks~maps~stack-fit*).

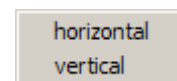
This is an excellent method to analyze and visualize chemical composition along a line. Quantitative thickness along the line can be generated if quantitative reference spectra (OD1) are used.

locate line – draws the (x,y) position of a linescan on the image currently displayed using information from the parameter file for the linescan (Jan2023: only set up for SDF format data written by STXM_control). Be sure to load and display the image on which the linescan was defined (or at least one which includes the spatial region of the linescan).

normalize to Io – divides each **horizontal** line by user-selected buffer containing the Io, then computes $-\log(\text{Image}/\text{Io})$. Get Io from *Add lines~Horizontal* if there is an open region in the linescan or from a separately recorded point or line spectral scan.

normalize to line

- Horizontal** - divides each **horizontal** line by content of user-selected buffer
- Vertical** - divides each **vertical** line by content of user-selected buffer



Hint: To remove line-by-line periodic scan noise, generate a 1-d profile of periodic noise by *Add lines~vertical* (or *Add lines~horizontal*, as appropriate) over all or part of an image then subtract that profile from the image using *Normalize to line*.

subtract reference computes (Image – ref), The reference 1-d profile signal is taken from user selected buffer (this routine does the same thing as *normalize to line ~horizontal*)

SPECTRA

Procedures for processing 1-d data (y as a function of x). This is often spectra, but can also be intensity profiles along a line, I₀ signals, etc

Absolute value – computes absolute value

Add

Append - append 2 data sets - all data points in overlapping region are kept

Buffer - generate SUM of 2 spectra (interpolation used)

Constant - add (or subtract) constant to y-axis

Calibrate (for each of **X** and **Y** axes):

Auto - uses last axis recalibration parameters

1 point – shift values by user defined amount from cursor selected point (shift)

2 point – shift values based on 2 points (shift & stretch)

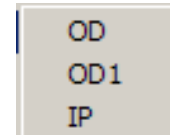
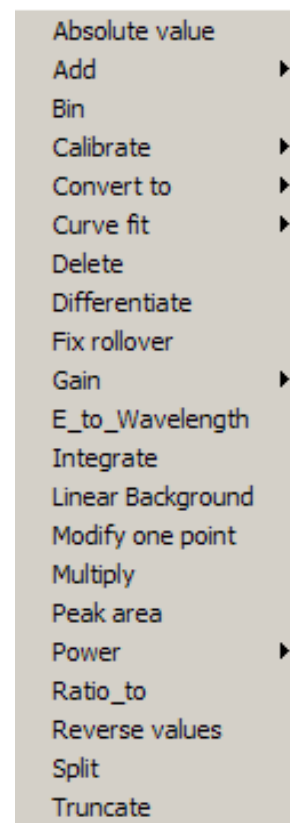
Numerical 1- or 2-point with numerical input

Convert_to

OD – automatically generate OD spectrum from a transmission spectrum by identifying a second buffer with I₀

OD1 – converts to intensity for 1 nm of material (needs XX-sf.od1)

IP – converts E-scale from kinetic energy to binding energy with user supplied photon energy. $IP = E_{\text{photon}} - E_{\text{kinetic}}$



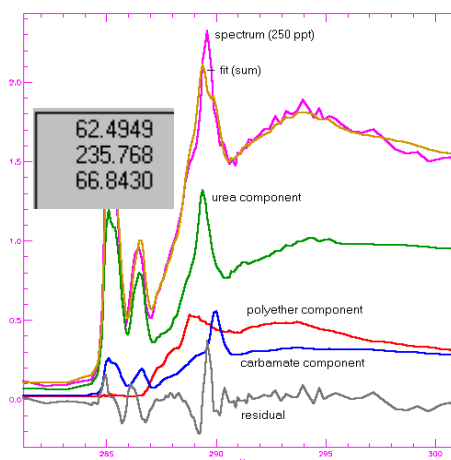
Curve fit

linear regression – use IDL regress function to fit the spectrum in the current buffer to a set of reference spectra. The reference spectra can be identified using a *.par parameter file (identical to that used in *stacks~map* and *linscan~fit*)

CGO fit - use conjugate gradient optimization method (CGO) to fit the spectrum currently displayed in a buffer to a set of reference spectra which the user is prompted to select from the disk. This is a useful method to check and display the fits that are obtained from image stacks. The fit components are listed on the IDL log as well as in the aXis2000 log display.

Pol fit – fit an angular dependent signal to

$$I(\theta) = C + A * \cos^2(\theta - B)$$



Spectra *(continued)*

Delete - delete all data between 2 cursor-identified positions

Differentiate – take derivative of displayed signal (simple $\Delta Y/\Delta X$ only)

Fix rollover – identifies data with modulo $2^{16}-1$ range, and adds units of to successive rolled over portions of the spectra

Gain - **multiply** – multiply y-axis values by a factor
- **divide** – divide y-axis values by a factor

E_to_wavelength - X-axis transformed by $12398/X$ (both way conversion)

Integrate – determine integral of displayed signal

Linear background - subtract user defined line

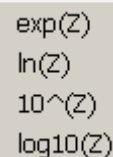
Modify one point – left click – add a point, right click – delete the point

Multiply – multiply the spectrum in the current buffer with that from a used selected buffer.

Peak area – determine area under curve between user-selected limits

Power

- **exp(Z)** – exponential of z
- **ln(Z)** - natural log of z-values
(use to convert Transmittance to Absorbance)
- **10^(Z)** – raise z-values to 10
- **log10(Z)** - base-10 log of z-values



exp(Z)
ln(Z)
10^(Z)
log10(Z)

Ratio_to - takes ratio of 2 buffers (interpolated to same x-axis scale)

Reverse values – reverses the (x) values in a spectrum.

Split – separates a multi-valued spectrum into single-valued regions, placed in successive buffers. Data of this type can be generated by acquiring multiple regions which are not in strict increasing-energy order. This can be useful as a means to track radiation damage. Recording the most chemical sensitive part of a NEXAFS spectrum at the beginning and end of a point spectrum is an effective way of having an internal check on radiation damage.

Hint: AXIS auto-detects multi-section spectra recorded with overlapping multiple regions and plots all components in the same buffer. The separate single-valued sections are placed in sequential buffers by this command.

Truncate - truncate spectrum to data between 2 cursor-identified positions

DISPLAY

Over Plot - display multiple spectra

No Rescale - select multiple buffers (0-9)

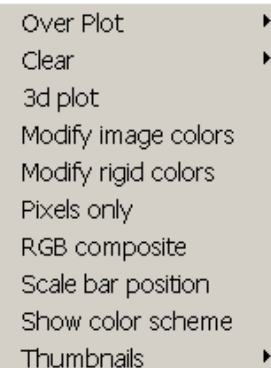
Y-axis of data is preserved

Rescale – select multiple buffers (0-9)

each spectrum is rescaled to full screen

Window - add a single buffer, with data rescaled within y-limits selected by the user using the cursor

Shift - add a single buffer → bottom y-position selected by cursor; no rescale (ie y-scale is set by previous data)



All modes of *Display~Over Plot* can be combined in any order. All previous *Display~Over Plot* processing is preserved in the main plot window until a single buffer is selected for plotting, by left-clicking on: thumbnail plot, buffer label, or indicator box.

Clear Lines – removes cursor-related lines from Main Image

Current – erases only the currently selected buffer

Selected - any of 0-9 buffers can be erased (also as second row button)

All - resets all buffers to zero (like starting a new version of aXis2000)

NB the **Clear Buffer** single line command accesses the last 2 commands

Hint: If the colors go 'crazy', restore the default color scheme by **Clear Buffer** or *Display~Clear*.

This often occurs on the first use of *stacks~analyze~zimba* – a black display is generated when displaying the first selected spectrum. Use the **Clear Buffer** command above the thumbnails to reset the color scale, and then erase and re-select the regions in *stacks~analyze~zimba*

3d-plot - generate 3a -d shade surface plot from an image with x,y,z axes.

Modify image colors - pop-up widget (XLOADCT) that selects color scheme and adjusts (top, bottom, gamma) variables. *In Win systems, if 256 colors is set, this updates all graphic windows dynamically. If color is set to higher value (16-bit, 24-bit, or true color) one needs to redisplay a graph to change the color scaling. Since each thumbnail sketch (upper left of screen) is displayed independently, the color scale of the thumbnail sketches will only change after they are selected.*

Modify rigid colors - select custom colors for buffer specific spectral colors, background, foreground, etc

Pixels only - plot using 1 display pixel per pixel of data

RGB composite – takes images in 3 buffers and assigns them to red, green and blue color components of a single image. It is necessary that all images are the same size and are of the same physical region. The composite image is saved to disk in a 3-component TIF format which reads into Paint Shop and Powerpoint. The individual R, B, G components can be read back using the *Read~Images~Other~TIF~data* command.

Display~ RGB composite / *continued*

User is given option to use a common scale for all three images (thus preserving information about relative intensities of components), or autoscaling each image independently, which will give equal visibility to each component.

Scale bar position – use cursor to define the left end of the scale bar.

Show color scheme - display current colors assigned. Note that the bottom 16 colors in the 256-color table are assigned to 'hard' colors to allow ready differentiation of different buffers.

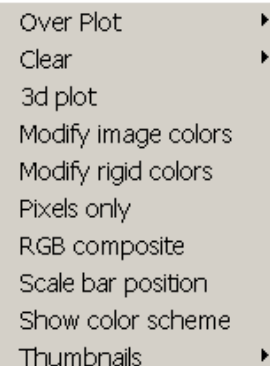
Thumbnails – display multiple buffers on main window

4 – user selects any 4 buffers

- **common scale** - plot using identical scale for all images (in B/W for printing)
- **rescale each** – plot with each image byte scaled to its data values (in color)

9 – display all 9 buffers

- **common scale** - plot in B/W (for printing)
- rescale each** – plot with each image byte scaled to its data values (in color)

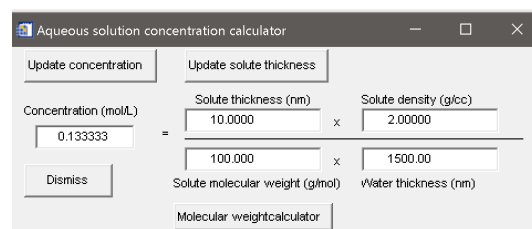
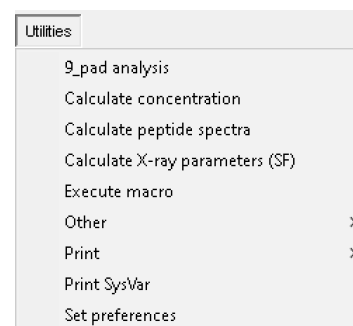


[return to top menu](#)

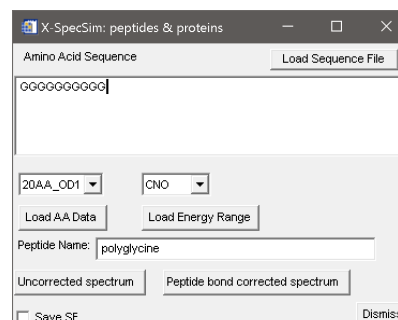
UTILITIES

9_pad analysis automated analysis of 9-pad radiation damage patterns. Templates for 10 areas are adapted to the spatial scale of an experimental 9-pad image, multiplied and integrated to generate the signal as a function of dose.

Calculate concentration computes equivalent concentration in aqueous solution (in Molarity). Use with *utilities~calculate X-ray absorption parameters (SF)* to ESTIMATE visibility of spectral features in aqueous solution, and estimate the solution concentration and water thickness to guide sample preparation. [see: Axis2000-manual-calc_conc.ppt 04-Jun-18]



Calculate peptide spectra – launches a widget to generate the NEXAFS spectrum of a user specified amino acid sequence from the C, N, O spectra of the constituent amino acids. Note, the AA sequence can be taken from the protein data bank (PDB) or generated from standard codes for the amino acids.



Utilities (continued)

Calculate X-ray parameters (SF) - Determine the **mass absorption** or **transmission** for user defined elemental compositions. The mass absorption elemental signal for a given chemical formula and density provides normalization for reference spectra to the quantitative OD1 (optical density response for 1 nm thickness) which can be used in the SVD map and stack_fit routines to obtain quantitative thickness maps. In addition, the transmission for a user-defined composition, density and thickness can be useful to evaluate feasibility of a sample. NB The *sf.pro* program (in the AXIS directory) is a full implementation of the old CXRO SF program for X-ray constants, which corrected some errors. Routine written by Billy Loo, 1996. The elemental X-ray mass absorption coefficients are from 1996. There may have been updates since then. See https://henke.lbl.gov/optical_constants/ for latest values.

Execute macro – execute a file of standard aXis2000 commands (with parameters) So far only a few of the axis commands are set up to be used in this way.

Other – commands not directly related to spectromicroscopy

ECHEM plot – generate composites of images and electrochemical results (e.g. cyclic voltammogram) to make a movie of in situ electrochemical results

ECHEM plot
ISEELS Display
print ISEELS Database

ISEELS Display – tool to plot spectra from an inner shell electron energy loss spectrometer.

print ISEELS Database – print all selected ISEELS spectra from the gas phase core excitation data base (<http://unicorn.mcmaster.ca/corex/cedb-title.html>)

Print

Logbook (default: 3" x 3") – no annotation

Annotated (default: 4" x 4") – brings up the IDL annotate widget.

Logbook
Annotated

*Hint: (1) A fast alternative to printing is to use a screen capture program such as **MWSnap** <http://www.mirekw.com/> to copy the screen area you want to the clipboard, then use an image processing program such as [Paint.net](http://www.paint.net/) to further modify (e.g. annotate). The modified screen captured image can then be transferred into a presentation program, such as powerpoint or keynote.*

*Hint: (2) Use The PRINTER option in *axis_XXXX.ini* (XXXX = win, unix, macos) to set up your default system printer*

Print Sysvar - display on the IDL log window (so, need licensed IDL) the current values of all IDL graphics parameters (used in code development).

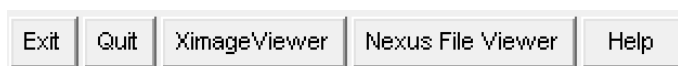
Utilities (continued)

Set preferences – allows user to modify the default parameters which are contained in a file called **axis.ini**. This uses a widget (*see fig*) to let the user define a large number of different parameters. The modified initialization file is written at the end of each normal exit to aXis2000 so that it starts in the same configuration (default directories etc) as in the last use.

On start-up, if AXIS cannot find the default initialization file (**axis.ini** in the subdirectory where the AXIS code files are located), it starts this widget.

Second row of single command buttons (A)

Above the Main graphical window



Exit write [axis.ini file](#) with parameters then return to main IDL command window. It is a good idea to use Exit rather than Quit, since Exit saves the last path used, which can save time navigating to a folder you were processing, the next time you launch aXis2000.

QUIT return to main IDL command window, without saving current parameters

NOTES:

If you have a net connection, and you have the command to start your browser identified correctly in the **axis.ini** file, then you can click on the indicated synchrotron sites or the aXis2000 label to connect to the X-ray microscopy facility. Clicking the label at the bottom of the splash screen takes you to the [aXis2000 web site](#)

Additionally, for those using a licensed version of IDL.

1. From the Command Window of IDL type *axis2000* to (re-)start AXIS.
2. If you wish to display the **rotating AXIS logo**, restart AXIS by typing **axis2000, /spin** at the IDL prompt.

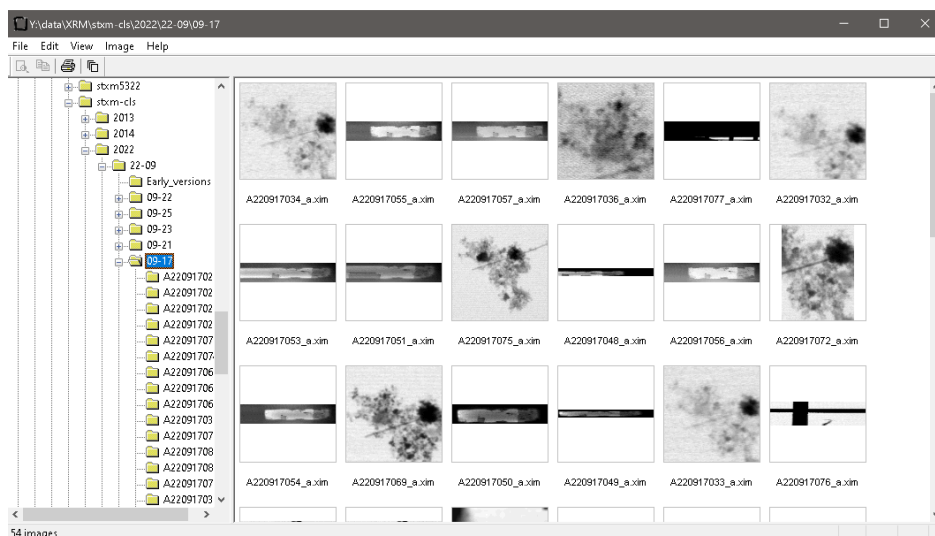
.

Second row of single command buttons (A) / continued

XimageViewer

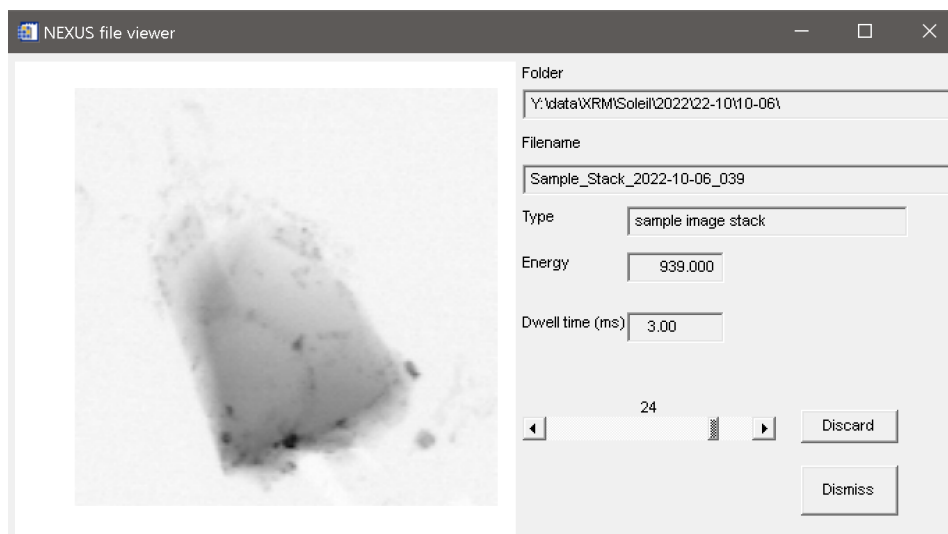
Launch a viewer of image files. In addition to standard image files (jpg, bmp, gif, tif, png, etc) this displays ALS_STXM (SDF) format file (*.hdf,*.xim) pairs in the selected folder. This is useful when you do not have detailed information on data files.

This DLL was provided by **Ivo Koprinarov**.



Nexus File Viewer

Launch a tool to review NeXus format files to allow rapid discard of unwanted files (the STXMs using pixelator acquisition save all files, 30-50% are junk). This is also useful to identify data of interest when the log book notes are insufficient. The data viewed in NEXUS file viewer is the default file name in *read~STXM (NeXus)*

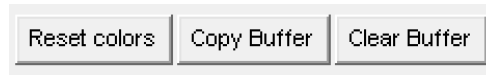


Second row of single command buttons (A)~NeXus file viewer / continued

Calls Adobe Acrobat reader, using the Unix (Acroread) or Windows command (AcroEx32) to bring up **AXIS.pdf** (a pdf version of the **aXis2000-manual.doc**). Since the location of the Adobe reader depends on the system, the system command that will start the Acrobat reader and display the AXIS.PDF file is part of the AXIS.INI file. These details are set by the **help_cmd** line in the **set_preferences** dialog, or by using a text editor to edit, axis-win.ini (Windows) or axis-unix.ini (unix, MacOS) file.

Second row of single command buttons (B)

above the 9-thumbail display



Reset colors

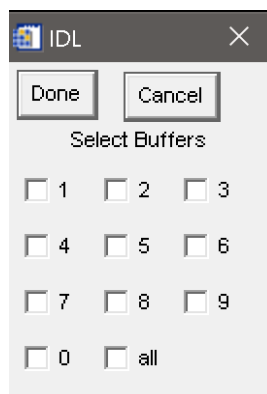
Changes Display color scheme to the current default. Often the color scheme is distorted after running one of the **stack~analyze** widgets. Use this button to reset to the default color scheme to that for aXis2000.

COPY buffer

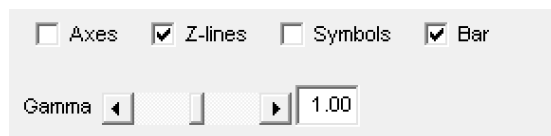
- moves contents of the current buffer to one of the 10 data buffers in AXIS. The destination buffer is selected by clicking on the thumbnail, or by clicking on the label of the buffer to which you wish to move the data. Note the latter is the only way to move data into buffer 0.

***Hint:** To preserve a processed spectrum or image located in Buffer 0 you must use Copy to transfer it to buffer 1-9 to avoid overwriting the result in the next processing step.*

CLEAR buffer pops up the multi-buffer selector widget to let the user select those buffers they wish to clear.



CONTROLS BELOW THE MAIN IMAGE WINDOW



Axes

If selected (**ON**), the X and Y scales are displayed.

If not selected (**OFF**, the default) only the image, X-axis and Y-axis labels are displayed.

Z-lines

- **ON** (default) - X, Y lineouts generated at every mouse move
- **OFF** - X, Y lineouts generated only on left mouse click

Note: The operation of *Zoom~pan* is also switched with this button. If ON, the zoomed image updates on every mouse motion. If OFF, the zoomed image updates only after a mouse click on the main image

Symbols

- ON** - plot symbols on lineouts and spectra (main image window only)
- OFF** (default) – no symbols on lineouts or spectra

Note these options allow one to adapt to the speed of the computer. Continuous updating of the lineouts and the zoom/pan, as well as symbol plotting, take large amounts of cpu/graphics resources and can slow aXis2000.

Bar

- ON** (default) display white scale bar [size is (1,2,5,10 pattern; within 5-10% of image]
- The position of the scale bar can be defined using *Display~scale bar position*
- OFF** – scale bar and value are not displayed

Gamma

use either the slider or the number box to change the image gamma (for the aficionados, see https://en.wikipedia.org/wiki/Gamma_correction). Note this control is only active if an image is displayed.

***** XXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXX*****

INSTALLATION

***** XXXXXXXXXXXXXXXXXXXXXXXXXXXXXXXX*****

Copy all code into a directory. (use **WinZip** (or similar) to **expand axis2000.zip** using **folder names** into your source directory). –

I STRONGLY RECOMMEND using c:\aXis2000\ on Windows systems, and users\aXis2000 on MacOS systems..



There should be 4 folders (AAData, java, logo, and sfdata) and over 500 files (mostly *.pro).

To set-up a **licensed IDL system** so it starts aXis2000 every time:

1. Start IDL (I use version 6.3 for Windows and 8.8 for MacOS)
2. Set up preferences
 - Path – add the directory with aXis2000 files
 - Start-up- working directory: c:\axis2000\ (or whatever your source directory is)
 - start-up file: axis2000_batch
3. Shut down IDL and restart.

To use IDL without automatically starting axis2000, type **aXis2000** at the IDL command line.

Hint1: There are often problems if you try to use a different drive letter, or folder name than c:\aXis2000. A particularly **BAD CHOICE** would be C:\Documents and Settings\Default User\aXis2000\ since the IDL code in aXis2000 automatically **TRUNCATES text strings at the first space. Thus the program would be looking for files (or writing files to) C:\Documents**, a folder which does not exist.

Hint2: I may be possible to execute aXis2000 just from the aXis2000.sav, compiled version. However, many parts of aXis2000 would not work properly as it needs information from the distributed source code.

Several different versions of the [INI file](#) are included: axis_win.ini (for Microsoft Windows); axis_macos.ini (for Apple OS), axis_unix.ini (for unix OS). Use the one identified for your operating system. If necessary use a text editor to edit the entries for your needs. Typically, this will involve changes to the SIZE parameter (I choose a value that fills about ½ of my graphics screen), the location of your pdf viewer, and the location of your web browser. See below for an example and more details.

The **aXis2000 initialization file** (axis_win.ini, axis_unix.ini, axis_macos.ini)

A sample AXIS.INI file (on a WIN system):

```
LINE#
1. c:\aXis2000\
2. WIN
3. PS
4. Y:\Backup\papers-old\X-ray_microscopy\CNTs\MWCNT-Najafi\
5. 1.00000
6. 0
7. mac
8. idl.ps
9. Arial*14
10. copy idl.win lpt1:
11. c:\progra~1\acrobat3\reader\acroRd32 c:\axis\axis.pdf
12. C:\Program Files (x86)\Google\Chrome\Application\chrome.exe
-----
written: Mon Jan 02 14:58:04 2023

; ***** Initialization file for AXIS Widget *****
;
; (aph 31-Jan-00) Please adapt to your hardware
; Items set                devices currently supported
; -----
1 CodePath                (e.g. c:\aXis2000\) - include final separator
2 Screen display device   (WIN, X)
3 Print file format        (PRINTERPCL, PS) ; Printer uses system default
4 Default data directory  (any - last path will be saved on Exit)
5 graphics scale factor   (0.5 - 2) (1 = 360x360 pixels for image window)
6 default color table      (any IDL supports) 0 = B/W; 3 = red temperature
7 location                 (NSLS has special meaning = spooler switch)
8 spooler or print file    (c:\tmp\xla at nsls - special !!)
9 default font for widget  (windows - Arial*14; unix - 6x10 or 8x13 ) NOT USED
10 Printer command line    (command line needed to transfer to printer)
                        NOT used if PRINTER is set on line 2
11 Command to view Help pdf (get acrobat reader free from www.adobe.com)
12 command to run browser   (WIN: must be executable from MS-DOS window)
```

A. For WINDOWS machines

Color schemes greater than 256 work fine. In most cases, data is redisplayed, but occasionally you need to refresh a display to get a new color table to be implemented (With color tables above 256 - 16-bit, 32-bit or true color) IDL does not automatically update the display). *Reset colors* does this.

B. For Unix (X-windows) based systems

Since Unix is case sensitive, it is important that the case of the names of files be preserved. All the AXIS file names are strictly LOWER CASE. Some FTP programs force the file names to upper case which is a problem. If you have error messages saying a given pro file cannot be found please check the case of the filename and rename it to exactly what the IDL error reports.

Use ASCII transfer for all files (except *axis.bmp*, *axis.doc*, *axis.pdf* associated with the stack codes, which must be transferred in binary mode) when transferring code from Windows to UNIX or Macintosh systems to avoid problems with the differences in end-of-line coding in the 3 systems.

- the UNIX version of IDL only likes @bsif_com or @axis_com in the AXIS.PRO file. On the Linux-PC version the batch processing would not work when long names such as axis_common.pro are used for include files. Some examples of the COMMON include files are:

axis_com.pro	ax_peem_com.pro
bsif_com.pro	img_com.pro

in total, there are 39 common files in aXis2000 (as of Jan 2023)

Run time hints:

*If you quit AXIS but stay in IDL, restart AXIS by executing **axis2000** at the IDL command line.*

IDL Virtual machine version

This allows you to run aXis2000 without purchasing an IDL license :

1. download IDL from the download website, which, as of January 20203 is:
<https://www.l3harrisgeospatial.com/Software-Technology/IDL>
(NB you need to register in order to access the download the code (version 8.6 is ~2 Gb)
2. Download the aXis2000.zip package from unicorn.mcmaster.ca/aXis2000.html
3. Extract (unzip) all contents into c:\aXis2000 (Windows) or /Users/aXis2000 (Mac OS X)
(Make sure 'Use folder names' is turned on when you unzip)
4. set up a **desktop icon** for convenience. If it is your first time setting up aXis2000, you need to associate the aXis2000.sav file with the IDL runtime engine which on windows is
(IDL6.3) C:\RSI\IDL63\bin\bin.x86\idltrt.exe
(IDL8.6) C:\Program Files\Harris\IDL86\bin\bin.x86_64\idltrt.exe (for 64-bit computers)
or C:\Program Files\Harris\IDL86\bin\bin.x86\idltrt.exe (for 32-bit computers)
Once the association of *.sav files with IDLRT is made the desktop icon can be created by
(Windows) right click on the desktop; click New~shortcut; browse to, then click on
C:\aXis2000\axis2000.sav; click Next; define name of shortcut.
5. If you have other *.sav files you do NOT want to associate with IDLRT, you can define the desktop item as outlined at <http://unicorn.mcmaster.ca/axis/aXis2000-IDLVM.html>

Hints:

You must have all the source code files in the folder to make the Virtual Machine version work properly

Note: If you use a folder other than c:\aXis2000 or /Users/aXis2000 there may be problems running the code.

Do NOT use folder (directory) names with blanks in the folder or file names for the source code or your data !!!

**A common problem is to put aXis2000 in 'My Documents' or in 'Program Files'
Note that both of those folder names contain a blank. You are warned.**

Please provide feedback by email on how you would like to see this program evolve to be more useful to the X-ray SpectroMicroscopy community.

If you report a bug, I will try to fix it as soon as possible. It is most useful if you can send data file(s) which exhibit the bug, and, if you are running a full version of IDL, the IDL log window error report.

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FOR BEGINNING USERS, a number of **USEFUL DOCUMENTS** are available by download from <http://unicorn.mcmaster.ca/aXis2000.html>

TUTORIALS – SIMPLE describes unit operations for manipulating X-ray microscopy data.
- **ADVANCED** - walks through a STXM data analysis using curve fitting

STACK_ANALYZE - manual to walk-through a typical stack analysis based on ALS STXM or PEEM data. *[Warning – this document is quite dated and oriented to the analysis of PEEM data measured at ALS_PEEM2 (on the 7.1 bend magnet beamline)]*

STACK_FIT - manual to walk through stack fit from a prepared *.ncb aligned image sequence.

FOR PROGRAMMERS – SOME USEFUL ADD-INS (in aXis2000.zip)

Content.lst - lists all AXIS routines with a one-line description

Run **ax_make_html** at the IDL prompt. This will extract the header documentation from all the files to generate an html document with internal hyper-links etc. (there is an HTML file in the aXis2000 distribution with this output)

TEST-DATA – a set of files that can be used to test each of the aXis2000 routines
(*contact Adam Hitchcock to obtain*)

GLOSSARY of terms associated with aXis2000

axis_XXX.ini - (XXX=win, unix, macOS) text file containing default parameters for aXis2000. Edit to adapt to your system

component maps - spatial distributions of a chemical species, which can be generated from multiple images (selected energies or a full image sequence) using *stacks~maps~SVD* or *stacks~maps~stack fit*)

dark count – signal (image or spectral) recorded without X-rays. This is detector specific (e.g. CCD camera leakage, light leakage into PMT etc) and must be subtracted from real signal before any data processing involving ratios (yield or absorption determinations).

IDL - Interactive Data Language is a high performance scientific computing platform, optimized for arbitrary sized multi-dimension array processing and image display. IDL was originally developed by David Stern and his team from Research Systems Inc. Wikipedia has [a short history of IDL](#). The current owner is [L3Harris](#)

IDL Virtual Machine – a version of IDL which allows execution of compiled IDL script. This can be downloaded for free from [L3Harris](#)

IDL widget - a graphical user interface with pre-programmed data manipulation or other capabilities written in Interactive Data Language.

ini file - a text file which contains values of parameters used to customize aXis2000 for your environment

intensity profile - Plot of pixel value across a line defined on an image. Rectilinear profiles are available automatically, updated either on each mouse move, or after a right click, depending on the setting of the continuous lineouts control.

line scan spectra – a 2-d data set in which the intensity along a line is recorded at a series of photon energies. aXis2000 displays the data with energy along the horizontal axis and position along the line as the vertical axis. Thus horizontal lineouts (lower side panel) display spectra at a point; vertical lineouts (left side panel) display the contrast along the physical line at a given photon energy.

masked image - Output of *Images~generate_mask*. It is an image consisting of only 0 or 1 value pixels, based on whether the original pixel was above (1) or below (0) a user-defined threshold. This, along with *Images~multiply buffers* and *Images~average pixels*, is useful to evaluate the intensity in a selected region of a component map.

netCDF – a standard binary scientific data format. The data is read and written by platform dependent routines provided by a standards body. Used for NSLS STXM data.

OD – optical density, or absorbance. In a transmission measurement (STXM, TXM) the recorded data is the intensity of the transmitted X-rays. It is converted to OD by $-\ln(I/I_0)$ where I is the signal transmitted through the sample and I_0 is the signal without the sample in place.

self defining files - A structured, ascii format in which a syntax is used to allow a single program to read (and write) complex data structures without prior knowledge of the data structure. Read and write routines are available in C++ and IDL.

Stack – a set of images at a sequence of energies (in other spectromicroscopies, sometimes called a ‘map’)

ZSTACK

Data Analysis for Hyperspectral X-ray Microscopy Imaging
v2.1 (5 feb 2001)

ZSTACK is a suite of IDL procedures for alignment and analysis of a series of x-ray microscopy images that have been acquired at different x-ray energies. It operates on Windows, Macintosh, and UNIX platforms using IDL v5.0 or later.

ZSTACK was written originally to satisfy my own needs for analysis of STXM spectral image stacks. The original version was based upon the original STACK code developed by Chris Jacobsen (SUNY - Stony Brook). It has expanded considerably beyond the initial scope, largely due to my analysis needs and feedback from fellow users.

In a collaborative effort with Adam Hitchcock (McMaster University), ZSTACK has been bundled into his AXIS data analysis package. ZSTACK can also be used independently of AXIS.

The development of ZSTACK has been an evolutionary process. Please notify me of any bugs, problems, comments, or suggestions for improvements or new features.

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ZSTACK Buildlist

The screenshot shows the ZSTACK Buildlist application window. The interface includes a directory browser at the top, a central image display area, a list of data files on the right, and various control buttons and input fields at the bottom. Eight numbered callouts provide a step-by-step guide to the software's functionality.

- 1** Browse for desired directory/folder
- 2** Pull-down list of data files in current directory
- 3** Select which image is displayed and how it is displayed
- 4** Parameters of displayed image
- 5** Set current image as first file
- 6** Build list of data files From first file to last file
- 7** Save or retrieve list of files
- 8** List of data files is complete Go on to next step: aligning the images

Additional callouts describe specific features:

- Current directory/folder**: Points to the directory path field.
- Current image**: Points to the central image display area.
- List of data files selected for alignment and analysis**: Points to the file list on the right, noting that the highlighted file is displayed.
- Retrieve binary file of previous saved stack data**: Points to the binary filename input field.
- Number of images Currently in list**: Points to the 'Number of Files' counter at the bottom right.

The file list on the right contains the following data:

Filename	Energy (eV)	Position (X, Y)
05FEB022.NC	281.61	44.028 A
05FEB023.NC	282.60	43.873 A
05FEB024.NC	284.01	43.655 A
05FEB025.NC	284.12	43.639 A
05FEB026.NC	284.22	43.623 A
05FEB027.NC	284.31	43.609 A
05FEB028.NC	284.41	43.594 A
05FEB029.NC	284.51	43.578 A
05FEB030.NC	284.62	43.562 A
05FEB031.NC	284.72	43.547 A
05FEB032.NC	284.81	43.533 A
05FEB033.NC	284.91	43.517 A
05FEB034.NC	285.01	43.501 A
05FEB035.NC	285.12	43.486 A
05FEB036.NC	285.21	43.472 A
05FEB037.NC	285.31	43.456 A
05FEB038.NC	285.41	43.441 A
05FEB039.NC	285.52	43.425 A
05FEB040.NC	285.61	43.411 A
05FEB041.NC	285.71	43.395 A
05FEB042.NC	285.81	43.380 A
05FEB043.NC	285.92	43.364 A
05FEB044.NC	286.01	43.350 A
05FEB045.NC	286.11	43.335 A
05FEB046.NC	286.22	43.319 A
05FEB047.NC	286.31	43.305 A
05FEB048.NC	286.41	43.289 A
05FEB049.NC	286.51	43.274 A
05FEB050.NC	286.62	43.258 A

ZSTACK Align

(before alignment)

The screenshot shows the ZSTACK Align software window. It is divided into several sections:

- Align images using :** Radio buttons for ☒ Original data and ☐ Data as displayed.
- Reference image for alignment :** Radio buttons for ☒ Each Preceding Image, ☐ Each Following Image, and ☐ Constant Image. Below is a **Select file** button.
- Edge enhancement before alignment:** Radio buttons for ☐ Sobel, ☐ Roberts, and ☒ None.
- Cross-correlation determination :** Radio buttons for ☒ Correlation maximum and ☐ Center of mass. Below are input fields for **Maximum image shift (pixels):** 10, **Image shift threshold (pixels):** 0.01, and **Edgegauss smoothing (pixels):** 3.
- Buttons:** **Select a subregion for alignment**, **Reset subregion**, **Start auto-alignment**, **Skip alignment**, and **Align images manually**.
- Image Display Area:** Three panels labeled **STXM Image**, **Correlation Fn**, and **Shifted Image**. Below them is a dropdown menu showing **05FEB022_NC: 281.61 eV, 44.028 Å** and buttons for **Display Previous Image**, **Display Next Image**, **Play movie**, **Display Parameters**, and **Plot Parameters**.
- Alignment Shift File:** A text field for **Alignment Shift Filename (*.aln):** 05FEB022_093.aln, with **Save shifts**, **Read shifts**, and **Browse *.aln** buttons.

Numbered callouts (1, 2, 3) point to specific sections:

- 1:** Points to the **Align images using :** and **Reference image for alignment :** sections.
- 2:** Points to the **Select a subregion for alignment** button.
- 3:** Points to the **Start auto-alignment** button.

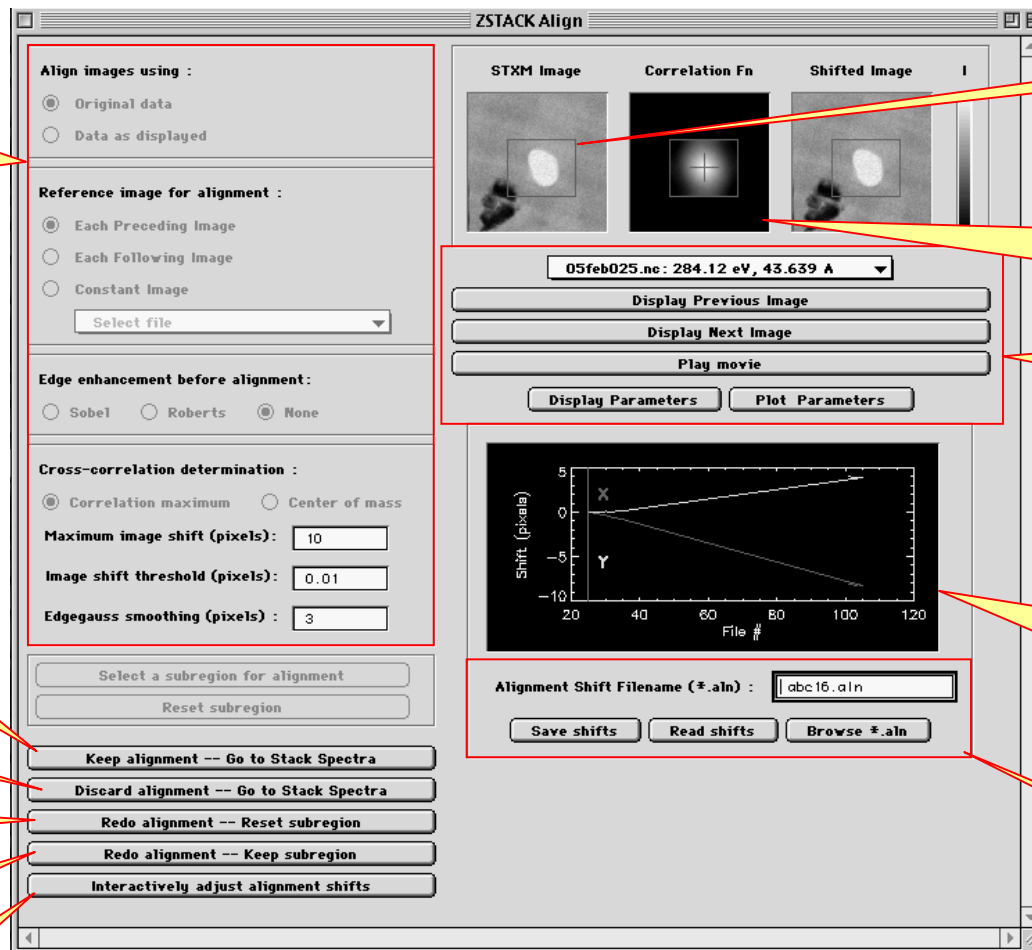
Other callouts describe various components:

- Choose the conditions for image alignment :** Reference image, Edge enhancement, Correlation determination.
- Pull-down list of data files** (points to the file selection dropdown).
- Define a sub-region in the image to use for alignment** (points to the subregion selection button).
- Start the auto-alignment of the images** (points to the Start auto-alignment button).
- Skip aligning the images Go on to extracting spectra** (points to the Skip alignment button).
- Align the images manually** (points to the Align images manually button).
- Raw STXM image** (points to the STXM Image panel).
- Color bar for images** (points to the color bar on the right).
- Select which image is displayed and how it is displayed** (points to the image display controls).
- Retrieve existing alignment shifts** (points to the Read shifts button).

After alignment shifts are retrieved from a saved file, this dialog window will be updated to show alignment shifts, alignment parameters, and shifted images

ZSTACK Align

(after alignment)



This now displays the conditions which were used to obtain current alignment

Subregion used for alignment

Correlation function crosshairs:
red for center of corr'n image
blue for maximum position
Red box is edge of corr'n image

Select which image is displayed and how it is displayed

Keep current alignment

Skip the alignment

Redo the alignment
erasing the subregion

Redo the alignment
keeping the subregion

Manually adjust
the alignment

Plot of alignment shifts
Cursor position corresponds
to current image and
can be moved by
clicking mouse in the plot

Save or retrieve
alignment shifts

ZSTACK Tune

The screenshot shows the ZSTACK Tune Alignment window. On the left, a control panel includes sliders for X and Y Alignment Shift, buttons for image navigation and parameter display, and options for alignment persistence. On the right, a 2x3 grid displays the STXM Image, Correlation Function, and Shifted Image. Below this is a Close-up Images section with a plot of alignment shifts over time.

Adjust the x shift using either the “New” text box or the slide bar

Adjust the y shift using either the “New” text box or the slide bar

Select which image is displayed and how it is displayed

Add fiducial points or shapes to close-up of shifted images

Keep new alignment

Keep old alignment

Reset to old alignment

Plot of alignment shifts
Cursor position corresponds to current image and can be moved by clicking mouse in the plot

By placing some fiducial points in the close-up of the shifted images, the quality of the alignment can be inspected while playing the images as a movie

ZSTACK Spectra

The screenshot shows the ZSTACK Spectra software interface. It includes a 'Select Regions for Spectra' panel on the left with buttons for adding and resetting regions. The main area displays 'Shifted Image' and 'Clipped Image' side-by-side. Below these are controls for selecting data files, saving spectra, and a plot of absorbance vs. energy (eV). Callout boxes provide detailed instructions for each major section of the software.

Define regions in image to extract spectra using Region of Interest dialog

If desired, retrieve an I0 spectrum (raw data, *.spc, *.xas)

Save extracted spectra as single beam, % transmittance, or absorbance in *.spc, *.xas, or *.gif format with all spectra in a single file or one file for each spectrum

Add fiducial points or shapes to close-up of shifted images

Images shifted after alignment

Shifted images clipped of edges where there is no longer any data due to shifting

Select which image is displayed and how it is displayed

Plot of spectra
Cursor position corresponds to current image and can be moved by clicking mouse in the plot

Get intensity vs position

Save images

Finished ? Quit here

Fourteen separate regions of interest can be specified, yielding fourteen I spectra.
In addition, a region of interest can be specified in an empty region of the sample to yield an I0 spectrum.

Color of the region of interest in the images corresponds to the same color spectrum

Region of Interest



ZSTACK Profile

The screenshot shows the 'Image Profile' window of the ZSTACK software. The interface is divided into several sections:

- Generate intensity profile using:** Includes radio buttons for 'Column (X)' and 'Row (Y)', both currently set to 50. Below these are instructions: 'YELLOW line is intensity profile', 'RED line is cursor', and 'Bottom Left corner is (0,0)'.
- Scale image intensity using:** Includes radio buttons for 'Intensity range of each image' (selected) and 'Intensity range of entire image stack'.
- Profile base filename:** A text field containing '05FEB022_093'.
- Save Profile Intensity as:** A button to save the profile.
- Spectra base filename:** A text field containing '05FEB022_093'.
- Save Spectra of Current Pixel as:** A button to save the current pixel's spectra.
- Save Spectra along Profile Line as:** A button to save spectra along the profile line.
- Buttons:** 'Display Previous Image', 'Display Next Image', 'Play Movie of Images', 'Display Parameters', 'Plot Parameters', 'Display Previous Profile', 'Display Next Profile', 'Play Movie of Profile', and 'Return to ZSTACK Spectra'.
- Image Viewers:** 'Shifted Image' and 'Profile Image' windows showing the current image with a yellow profile line and a red cursor.
- Plots:** Two plots are shown: 'Image Intensity' vs 'Row #' (0 to 100) and 'Absorbance' vs 'eV' (280 to 295).

Callout boxes provide additional information:

- Adjust position of cursor and profile axis:** Points to the 'Generate intensity profile using' section.
- Save intensity profile:** Points to the 'Save Profile Intensity as' button.
- Save spectra of current pixel:** Points to the 'Save Spectra of Current Pixel as' button.
- Save spectra of each pixel along profile axis:** Points to the 'Save Spectra along Profile Line as' button.
- Select which image is displayed and how it is displayed:** Points to the 'Display Previous Image', 'Display Next Image', and 'Play Movie of Images' buttons.
- Select which profile is displayed and how it is displayed:** Points to the 'Display Previous Profile', 'Display Next Profile', and 'Play Movie of Profile' buttons.
- Finished? Return to ZSTACK Spectra:** Points to the 'Return to ZSTACK Spectra' button.
- Plot of spectra:** Points to the 'Absorbance' vs 'eV' plot.
- Plot of intensity along profile axis:** Points to the 'Image Intensity' vs 'Row #' plot.
- Cursor position corresponds to current position along profile axis and can be moved by clicking mouse in the plot:** Points to the 'Image Intensity' vs 'Row #' plot.
- Map of intensity as a function of x-ray energy and position along the profile axis:** Points to the 'Profile Image' window.
- Cursor position and x-ray energy can be moved by clicking mouse in the plot:** Points to the 'Image Intensity' vs 'Row #' plot.
- Cursor position can be moved by clicking mouse in the image:** Points to the 'Shifted Image' window.
- Color of the region of interest in the images corresponds to the same color spectrum:** Points to the 'Absorbance' vs 'eV' plot.
- The profile axis is the yellow line in both the shifted image and the profile image. The intensity along this line is plotted as the profile intensity.** Points to the 'Image Intensity' vs 'Row #' plot.
- Red cursor in profile plot corresponds to position of red line in both the shifted image and the profile image.** Points to the 'Image Intensity' vs 'Row #' plot.

ZSTACK Save

Specify base filename and directory to save files

Select a subregion of image to save

Trim images to eliminate edges clipped off during alignment

Save images in a variety of formats, optionally including spectra, roi, and legend info

Save images as a movie, optionally including spectra, roi, and legend info

Save image stack as a binary data file

After specifying ALL the options above,
Click here to save everything (images, movies, binaries) at one time.

The ZSTACK Save dialog box contains the following sections and options:

- Base filename :** 05FEB022_093
- Directory :** Marvin HD:RSI:IDL 5.2:test:98_05FEB:
- Save subregion of data ?** ☐ Yes ☒ No
Select subregion **Reset subregion**
- Save data after clipping edges ?** ☒ Yes ☐ No
- Save Regions of Interest ?** ☐ Yes ☒ No
- Image format :**
 - ☐ JPEG ☐ GIF ☐ TIFF ☐ PNG
 - ☒ Current image ☐ All images ☐ Spectra
 - Orientation** (dropdown menu)
 - ☐ Filename ☐ X-ray Energy ☐ Scale bar
 - ☒ Spectral Regions of Interest ☐ Color bar
- Movie format :**
 - ☐ MPEG ☐ MGIF
 - ☒ Images ☐ Spectra
 - Orientation** (dropdown menu)
 - ☐ Filenames ☐ X-ray Energies ☐ Scale bar
 - ☒ Spectral Regions of Interest ☐ Color bar
- Binary data format :**
 - ☐ *.STK ☐ *.NCB ☐ *.HDF
 - ☐ Header file
- NB : Disabled buttons are features that are not fully implemented**
- Save** button

On the right side of the dialog box:

- Shifted Image** and **Clipped Image** preview windows.
- 05FEB022.NC: 281.61 eV, 44.028 Å** (dropdown menu)
- Display Previous Image**, **Display Next Image**, **Play movie**, **Display Parameters**, **Plot Parameters** buttons.
- Plot of spectra** showing Absorbance vs. eV (280 to 300). A cursor is positioned at approximately 285 eV.
- IDL Slicer** and **Return to ZSTACK Spectra** buttons.

Images shifted after alignment

Shifted images trimmed of edges clipped during alignment

Select which image is displayed and how it is displayed

Plot of spectra
Cursor position corresponds to current image and can be moved by clicking mouse in the plot

Color of the region of interest in the images corresponds to the same color spectrum

Process data using IDL Slicer3

Finished ?
Return to ZSTACK Spectra

ZSTACK Display

The screenshot shows the 'ZSTACK Display Parameters' window with several sections and controls:

- Image zoom factor:** A text input field set to 1.00.
- Movie delay (sec per frame):** A text input field set to 0.100.
- Closeup image zoom factor:** A text input field set to 4.0.
- Profile image zoom factor:** A text input field set to 1.0.
- Display image intensity using:** Radio buttons for 'Absolute' and 'Percentage' (selected).
- Display minimum:** A text input field set to 0.0.
- Display maximum:** A text input field set to 100.0.
- Display Gamma:** A text input field set to 1.00.
- Scale image intensity using:** Radio buttons for 'Intensity range of each image' (selected) and 'Intensity range of entire image stack'.
- Buttons:** 'Load New Color Table', 'Invert Color Table', and 'Select Plot Colors'.
- Display images as:** Radio buttons for 'Original data' (selected), 'Images / current image', '-log (images/current image)', 'Images - current image', 'Images / I0 spectrum', '-log (images / I0 spectrum) [Absorbance]', 'Images - I0 spectrum', and 'Current stack - reference stack'.
- Reference spectrum:** A 'Select' dropdown menu.
- Scale factor:** A text input field set to 1.0.
- Reference image:** A 'Select' dropdown menu.
- Scale factor:** A text input field set to 1.0.
- Reference stack:** A 'Select' dropdown menu.
- Scale factor:** A text input field set to 1.0.
- Image preview:** Two side-by-side grayscale images labeled 'Shifted Image' and 'Clipped Image'.
- Buttons:** 'Display Previous Image', 'Display Next Image', 'Play movie', and 'Plot Parameters'.
- Spectrum plot:** A line graph showing 'Absorbance' (y-axis, 0.0 to 2.5) vs 'eV' (x-axis, 280 to 300).
- Buttons:** 'Update Image Display' and 'Close without Update'.

Callouts provide additional information:

- Modify zoom Factors and movie play rate:** Points to the zoom factor and movie delay controls.
- Modify intensity range for images:** Points to the display minimum and maximum controls.
- Modify scale range for images:** Points to the scale image intensity using controls.
- Modify color scale for images:** Points to the color table buttons.
- Modify type of image displayed:** Points to the 'Display images as' radio buttons.
- Future feature: Define reference spectrum, Image, or image stack For subtraction or ratio:** Points to the reference selection dropdowns.
- Images shifted after alignment:** Points to the 'Shifted Image' preview.
- Shifted images trimmed of edges clipped during alignment:** Points to the 'Clipped Image' preview.
- Select which image is displayed and how it is displayed:** Points to the 'Display Previous Image', 'Display Next Image', and 'Play movie' buttons.
- Plot of spectra Cursor position corresponds to current image and can be moved by clicking mouse in the plot:** Points to the spectrum plot.
- Finished ? Update or Discard changes:** Points to the 'Update Image Display' and 'Close without Update' buttons.

ZSTACK Plot

The screenshot shows the 'ZSTACK Plot Parameters' dialog box. It is divided into several sections with various controls and a central display area.

- Image zoom factor:** A text input field set to 1.00.
- Movie delay (sec per frame):** A text input field set to 0.100.
- Spectrum Offset:** A text input field set to 0.00.
- Display spectra as:** Three radio buttons: 'Single beam', '% Transmittance', and 'Absorbance' (which is selected).
- Plot Scaling:** Two sections. The 'X Range' section has 'Autoscale' checked and input fields for Minimum (280.000) and Maximum (300.000). The 'Y Range' section has 'Autoscale' checked and input fields for Minimum (0.000) and Maximum (2.500).
- Color and Plot Options:** Three buttons: 'Load New Color Table', 'Invert Color Table', and 'Select Plot Colors'.
- Image Display:** Two side-by-side grayscale images labeled 'Shifted Image' and 'Clipped Image'. A vertical color bar is to the right of the 'Clipped Image'.
- Image Selection and Control:** A dropdown menu showing '05FEB022.NC: 281.61 eV, 44.028 Å'. Below it are buttons for 'Display Previous Image', 'Display Next Image', 'Play movie', and 'Display Parameters'.
- Spectral Plot:** A line graph showing 'Absorbance' on the y-axis (0.0 to 2.5) versus 'eV' on the x-axis (280 to 300). A vertical cursor is positioned at approximately 288 eV.
- Final Actions:** Two buttons at the bottom: 'Update Image Display' and 'Close without Update'.

Callouts provide additional context for these features:

- Modify zoom factor, movie play rate, spectrum offset:** Points to the zoom factor, movie delay, and spectrum offset input fields.
- Modify options to display spectra:** Points to the 'Display spectra as' radio buttons.
- Modify scale range for spectra:** Points to the 'Plot Scaling' section.
- Modify color scale for images AND plot colors:** Points to the color and plot options buttons.
- Images shifted after alignment:** Points to the 'Shifted Image'.
- Shifted images trimmed of edges clipped during alignment:** Points to the 'Clipped Image'.
- Select which image is displayed and how it is displayed:** Points to the image selection dropdown and control buttons.
- Plot of spectra. Cursor position corresponds to current image and can be moved by clicking mouse in the plot:** Points to the spectral plot and its cursor.
- Finished? Update or Discard changes:** Points to the 'Update Image Display' and 'Close without Update' buttons.