KARTHALA
two-photon microscopy system

USER GUIDE

Open Data Plugin

Fiji Plugin — AODscope System

Reference document — Opening Acquisitions in Fiji

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1. Purpose of this document

This document describes how to install and use the **Open data** plugin for Fiji. The plugin lists the snap and stack acquisitions of an experiment folder and opens the selected acquisitions in Fiji, with the pixel size read from the acquisition `.ini` file.

> **ℹ Where to find the plugin**
> Once installed, the plugin is available in the Fiji menu **Plugins > AODscope > Open data**.

2. Installation

1. Close Fiji if it is running.
2. Copy the file `AODscope_Open_Data.jar` into the `plugins` folder of your Fiji installation (for example `C:\Karthala system\Fiji.app\plugins`).
3. Start Fiji, or use **Help > Refresh Menus** if Fiji is already open.
4. Check that the entry **Plugins > AODscope > Open data** is present.

3. Expected data organization

The plugin looks for `snap` and `stack` sub-folders (upper or lower case) in the experiment folder. Each acquisition is a folder inside them, containing its images (`.tif` or `.raw`) and its `.ini` file:

```
2026_10_22/
|-- snap/
|   |-- snap_001/    (*pmt1*.tif, *pmt2*.tif + .ini)
|   `-- snap_002/    (*pmt1*.raw, *pmt2*.raw + .bat + .ini)
`-- stack/
    `-- stack_001/
```

The experiment folder can be either a **day folder** (as above) or a **parent folder** containing several day folders. In the second case, the name of the day folder is added at the beginning of each entry of the list.

> **⚠ Image file names**
> Only `.tif` / `.tiff` / `.raw` files whose name contains `pmt1` or `pmt2` are opened. Other files are ignored.

Raw files (.raw)

A `.raw` file has no header, so the size of its images is read from the `.bat` file found in the acquisition folder. The `.bat` file must contain the import parameters, for example:

```
image=8-bit width=102 height=102 number=210
```

- `image=` is the pixel type: `8-bit`, `16-bit Unsigned`, `16-bit Signed`, `32-bit Real`, `24-bit RGB`... A type with a space is written in square brackets, for example `image=[16-bit Unsigned]`. Without `image=`, 8-bit is assumed.
- `width=` and `height=` give the size of one image, in pixels.

- `number=` gives the number of images in the file (1 if absent).
- `offset=` and `gap=` (in bytes) and the keyword `little-endian` are optional.

If the `.bat` file has several lines with parameters, the line that contains the name of the `.raw` file is used; otherwise the first line is used.

> ⚠ **Raw files not opened**
> If a folder contains both `.tif` and `.raw` images, only the `.tif` images are opened. If the `.bat` file or its width / height is missing, or if the `.raw` file is smaller than the parameters announce, the file is not opened and the message “Raw files” lists the problem.

4. Opening procedure

1 Start the plugin

Click **Plugins > AODscope > Open data**. The plugin window opens with the list of the folder used last time. The first time, the list is empty and the message “No folder selected” is displayed (picture below).



Plugin window at start-up

2 Choose the experiment folder

Click **Open folder...** and select the experiment folder. The list is filled with all the snap and stack acquisitions found, and the window title shows the name of the folder. **Open folder...** can be used again at any time to switch to another experiment.

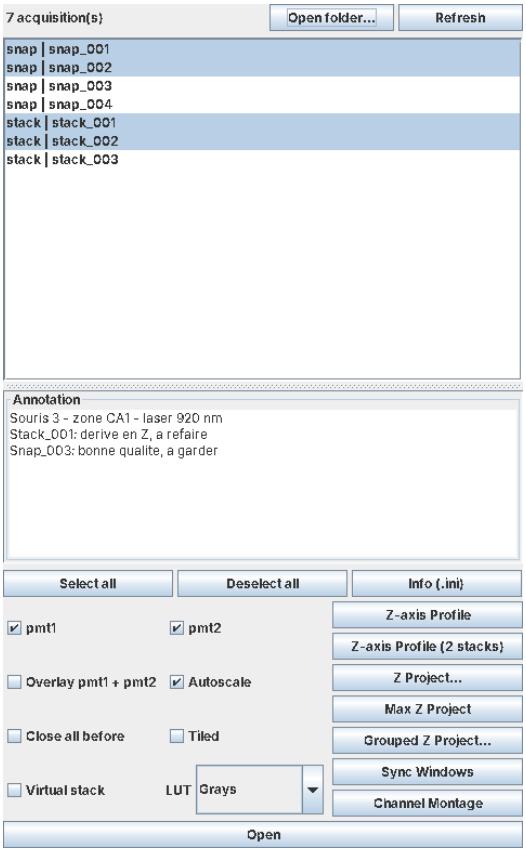
During an acquisition session, click **Refresh** to read the folder again. New acquisitions are added to the list, the current selection is kept, and the counter shows how many acquisitions are new.

If the experiment folder contains an annotation file named `<date>_annotation.txt` (for example `2026_10_21_annotation.txt`), its text is displayed in an **Annotation** area under the list, each time a

folder is loaded or **Refresh** is clicked. This area is shown only when an annotation file is present. When the folder contains several day folders, the annotation of each day is shown, one after the other.

3 Select the acquisitions

Click an acquisition to select it (a **double-click** opens it directly). Use **Ctrl + click** to add or remove an acquisition, and **Shift + click** to select a range. **Select all** and **Deselect all** act on the whole list.



Plugin window with the list of acquisitions (example data)

4 Choose the options

Tick the channels to open (**pmt1**, **pmt2**) and the display options (**Overlay**, **Autoscale**, **Close all before**, **Tiled**, **Virtual stack**, **LUT**). Each option is described in section 5.

5 Open the images

Click **Open**. For each selected acquisition, the pmt1 and/or pmt2 images (`.tif` or `.raw`) are opened with the pixel size applied. The window stays open, so that other selections can be opened afterwards.

> **ℹ No image found**
> If the selection contains no pmt1 / pmt2 `.tif` or `.raw` image, the message “No image” is displayed.

5. Buttons and options

Buttons

Button	Action
Open folder...	Chooses another experiment folder and reloads the list.
Refresh	Reads the current folder again to find new acquisitions. The selection is kept.
Select all	Selects all the acquisitions of the list.
Deselect all	Clears the selection.
Info (.ini)	Shows the content of the `.ini` file of each selected acquisition in a text window.
Open	Opens the selected acquisitions.
Z-axis Profile	Plots the Z-axis profile of the active image (see section 6).
Z-axis Profile (2 stacks)	Plots the Z profiles of two stacks on the same graph (see section 6).
Z Project...	Projects the active stack with the Fiji **Z Project...** command (see section 6).
Max Z Project	Maximum intensity projection of the active stack, in one click (see section 6).
Grouped Z Project...	Projects the active stack by groups of slices (see section 6).
Sync Windows	Opens the Fiji panel that synchronizes several image windows (see section 6).
Channel Montage	Puts pmt1, pmt2 and overlay of the active acquisition side by side in one image (see section 6).

Options

Option	Effect
pmt1 / pmt2	Opens the `.tif` (or `.raw`) images whose name contains `pmt1` and / or `pmt2`. Both are ticked by default.
Overlay pmt1 + pmt2	Creates an RGB image named `<acquisition>_overlay` (pmt1 in green, pmt2 in red). Both channels are opened, even if only one is ticked. The original pmt1 and pmt2 images are kept.
Autoscale	Applies an automatic contrast (0.35 % of saturated pixels) to the opened images. This changes the display only; the pixel values are not modified. Ticked by default.
Close all before	Closes all the images currently open in Fiji before opening the selection.
Tiled	After opening, arranges the image windows side by side (same as **Window > Tile**).
Virtual stack	Opens stacks as virtual stacks: the slices are read from the disk when they are displayed, which saves memory for large stacks. Works for `.tif` and `.raw` files. Unticked by default. Ignored when **Overlay** is ticked. With **Autoscale**, the contrast is adjusted on the displayed slice only. A compressed `.tif` is opened normally.
LUT	Lookup table of the opened images: **Grays** (default), **Grays (inverted)**, **Turbo**, **Viridis**, **Fire**, **Hot**, **Ice**, **Spectrum**, **Red**, **Green**, **Blue**, **Cyan**, **Magenta**, **Yellow** or **Green / Magenta** (pmt1 in green, pmt2 in magenta). The **Overlay** image is not changed. This changes the display only; the pixel values are not modified.

> **ℹ Settings remembered**
> The plugin remembers the last folder and the state of all the check boxes and of the **LUT** menu. They are restored the next time the plugin is started.

6. Tools for open images

The seven buttons on the right of the check boxes (**Z-axis Profile**, **Z-axis Profile (2 stacks)**, **Z Project...**, **Max Z Project**, **Grouped Z Project...**, **Sync Windows** and **Channel Montage**) act on the images that are already open in Fiji.

Z-axis Profile

Plots the mean intensity of each slice of the **active image** (the last image selected in Fiji). This is the same as **Image > Stacks > Plot Z-axis Profile**. If the image has a selection (rectangle, oval...), the profile is measured inside it; otherwise it is measured on the whole image.

Z-axis Profile (2 stacks)

Plots the Z profiles of two stacks on the same graph. A window asks for:

- **Stack 1** and **Stack 2**: the two open stacks to compare.

- **Use the ROI of stack 1 for both stacks**: ticked by default. Draw the ROI on stack 1 before clicking. If unticked, each stack uses its own selection (or the whole image if it has none).
- **Normalize**: divides each profile by the value of its first slice.
- **Show the table of values**: ticked by default.
- **Time interval between slices** and **Time unit**: with an interval of 0, the X axis is the slice number; otherwise it is the time, starting at 0.

The result is a graph named “Z profiles: stack 1 vs stack 2” (stack 1 in red, stack 2 in blue, with a legend) and, if ticked, a table named “Z profile values”.

Z Project...

Runs **Image > Stacks > Z Project...** on the **active stack**. The Fiji window that opens asks for the first and last slice and the **projection type** (for example Max Intensity, Average Intensity or Sum Slices). The result is a new image.

Max Z Project

Makes the **maximum intensity projection** of the active stack in one click, without any dialog. The result is a new image.

Grouped Z Project...

Runs **Image > Stacks > Tools > Grouped Z Project...** on the **active stack**. The Fiji window that opens asks for the **projection method** and the **group size** (number of slices projected together). The projection is then done for each group of slices of the stack.

Sync Windows

Opens the Fiji **Synchronize Windows** panel (**Analyze > Tools > Synchronize Windows**). It is used to synchronize several open image windows, so that an action in one window (for example changing the slice or moving the cursor) is also applied to the others. The synchronization is chosen in this panel, with its **Synchronize All** and **Unsynchronize All** buttons.

Channel Montage

Builds a new image named `<acquisition>_montage` with the **pmt1**, **pmt2** and **overlay** images of the active acquisition side by side, each one labelled. The images must already be open, at least two of them, with the same size. The montage uses the chosen LUT, the displayed contrast and the pixel size. For stacks, **all the slices** are processed and the result is a stack with the same number of slices (the stacks must have the same number of slices).

> **ℹ Messages**
> For the Z buttons and Max Z Project, the message “No image” is displayed if no image is open, and “Not a stack” if the active image has a single slice. The 2-stack profile needs at least two open images, and two different stacks.

> **⚠ Virtual stacks**
> On a virtual stack, a profile or a projection reads every slice from the disk, which can take some time.

7. Image calibration

For each acquisition, the plugin reads the `.ini` file found in its folder and applies the following values to the opened images (`.tif` and `.raw`):

- the pixel size in XY, from the line containing `pixel size (um)`, in µm;
- for stacks, the step in Z, from the line containing `increment Z`, in µm.

A decimal comma is accepted in the `.ini` file.

> ⚠ **Missing value**
> If the `.ini` file or the value is not found, the scale of the image is left unchanged. Check the scale (**Image > Properties...**) when in doubt.

8. Command-line launch

The plugin can be started from a terminal with the experiment folder already loaded:

```
"C:\Karthala system\Fiji.app\ImageJ-win64.exe" -eval "run('Open data',
'select=C:/Users/xx/xxx/xx/2026_10_22/ tiled');"
```

- `select=` gives the experiment folder. Use forward slashes (`/`) in the path.
- If the path contains spaces, put it in square brackets: `select=[C:/my data/2026_10_22/]`.
- The optional keyword `tiled` ticks the **Tiled** option at start-up.
- Without `select=`, the window opens on the folder used last time, or empty the first time (then choose the folder with **Open folder...**).

> ⚠ **Quotes**
> Inside the `-eval` text, use single quotes around the plugin name and the options. Double quotes would end the text given to the command line.

9. Quick reference

- Copy `AODscope_Open_Data.jar` into the Fiji `plugins` folder and restart Fiji.
- Start **Plugins > AODscope > Open data**; the first time, click **Open folder...**.
- Select the acquisitions (**Ctrl** / **Shift**) and choose the channels, options and **LUT**.
- Click **Open** or double-click an acquisition. Use **Refresh** to find new acquisitions.
- For `.raw` images, the `.bat` file must give `image=`, `width=`, `height=` and `number=`.
- Use **Z-axis Profile** on the active stack, or **Z-axis Profile (2 stacks)** to compare two stacks.
- Use **Max Z Project**, **Z Project...** or **Grouped Z Project...** to project the active stack.
- Use **Info (.ini)**, **Sync Windows** and **Channel Montage** to read settings, synchronize and compare.