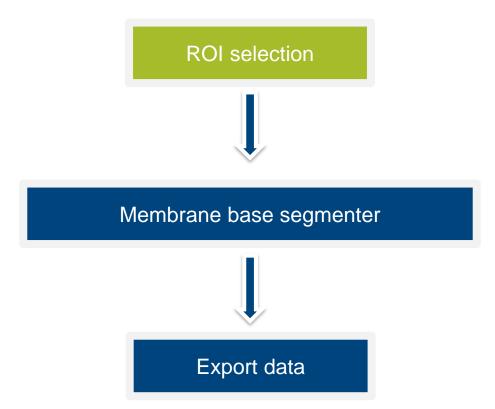




How to guide - Sample Pipeline «DETECT CELLS BASED ON MEMBRANES»

The pipeline purpose is to identify cell body starting from the membrane markers.

Working Flowchart





How to guide - Sample Pipeline «Detect Cells Based on Membranes»

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- 8. Modify the pipeline

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1. Download the demo dataset

Step 1

In order to run the pipeline described here below, please download the demo dataset according to the following instruction.

Step 1.1

Click on the below link to access to the Arivis downloading demo dataset's area.



arivisVision4D-DemoData-SamplePipelines-Membrane.zip file is saved on the download folder.



Step 1.2

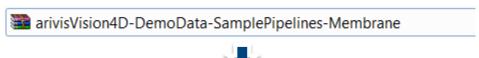
Create a new folder on your local disk.

Move the ZIP file from the download folder inside it.

Step 1.3

UnZip the file:

arivisVision4D-DemoData-SamplePipelines-Membrane.zip



Three files are now available in the folder.

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2. Open the demo dataset

Step 2

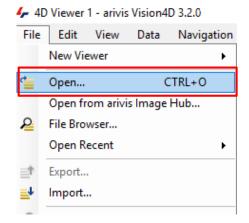
Open the Dataset on Vision4D.

Step 2.1

Select the *Open.*. item from the file menu.

Step 2.2

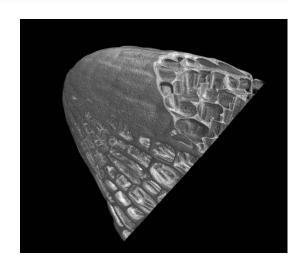
Select the dataset from the file browser.



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TIPS:

The dataset is visualized according to the current rendering setting parameters. Please refer to the (*arivis Vision4D Help*) for further details.



DETAILS:

The dataset is a multi dimensional, discrete, representation of your real sample volume. It can be structured as a Z series of planes (Optical sectioning) of multiple channels (dyes) in a temporal sequence of time points (located in several spatial positions).

Usually the dataset shows a single experimental situation (a complete experiment can be composed by several datasets). The datasets are available as graphic files saved in plenty of file formats (standard formats as well as proprietary formats)

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3. Select and activate the sample pipeline

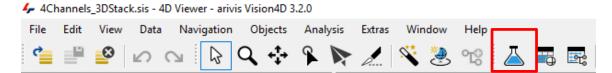
Step 3

Select and activate the ****Detect Cells Based on Membranes**** pipeline.

Step 3.1

Open the Analysis panel





Step 3.2

Drop down the analysis panel menu.



Step 3.3

Select Sample Pipeline.

Ready to run Detect Cells or Particles Analysis Pipeline 2020-01-08 New Pipeline... Sample Pipelines Duplicate... Rename...

Step 3.4

Click on the **Detect Cells Based**on **Membranes** item.

Detect Cells Based on Membranes

TIPS:

The active Pipeline, if any, will be replaced by the new one. Please refer to the (arivis Vision4D Help) for further details.



4. Pipeline operators layout

Step 4

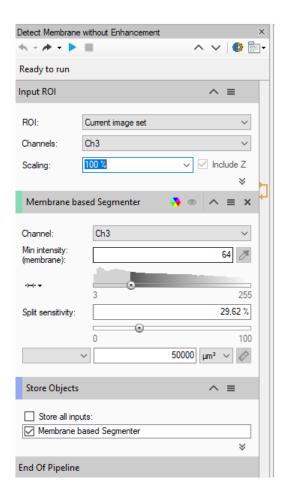
Pipeline operators layout.

Step 4.1 Region Of Interest: This operator allows the region of interest (ROI) selection. ROI defines the dataset subarea that will be processed and analyzed by the pipeline.

Step 4.2 *Membrane based segmenter:*

Detects the cell body starting from the membrane.

Step 4.3 *Store Objects*Store the detected segments (TAG) in the active dataset.





5. Execute the pipeline step by step

Step 5

Execute the pipeline step by step.

DETAILS:

The pipeline can be executed step by step (back and forth). This method allows to run and undo a single Operation. Either the arrow buttons or the Operation list can be used to go through the operators list.

Step 5.1

Run the single operator





Step 5.2 (optional)

Undo the single operator





TIPS:

Undo the last operator executed if you need to change the operator settings.

6 - Execute the pipeline in a single run.

Execute the pipeline in a single run.

Step 6.1

Run the whole pipeline





Step 6.2 (optional)

Stop the pipeline execution





DETAILS:

This icon, located on the right side of the operator title bar, shows the operator status.





Task running



Task completed



7. View the results

Step 7

View the results.

TIPS:

Results (segments and measurements) will be stored in the dataset only if the Store Objects operator has been correctly set.

Please tick appropriately the option as shown below before complete the pipeline execution.



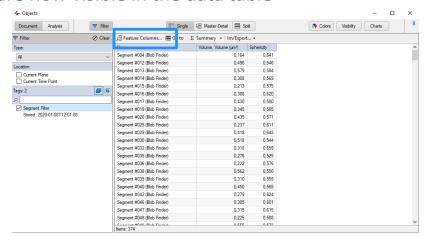
Step 7.1

Open the data table (if not already visible)





Measurements are now visible in the data table



TIPS:

Features can be added or removed from the data table using the *Feature Column* command.

Please refer to the (*arivis Vision4D Help*) for more details





Step 8

Modify the current pipeline.

DETAILS:

The pipeline can be modified to be adapted to another datasets. Therefore, all the pipeline parameters should be set according to the new dataset features.

Step 8.1

Switch the Viewing area from 4D to 2D view mode.



TIPS:

Before starting to modify the Pipeline layout, switch the Viewing area from 4D to 2D view mode.

During analysis setup, the Operator preview mode is only available in 2D mode. Once the pipeline has been executed, you can switch back to 4D view mode to display the segments. Please refer to the (**User Guide**) for more details

DETAILS:

Almost all the operators have the preview icon located on the panel header (title bar).

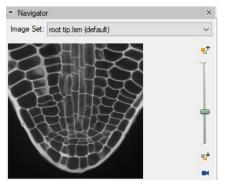




Click on this icon to preview the operator result on the current plane / time point

TIPS:

Use the Navigator Panel, located on the workspace area, to select the preview Z plane and/or Time Points (if any). Please refer to the (*arivis Vision4D Help*) for more details





Step 8.2

Change the Input ROI' operator parameters

Step 8.2.1 – ROI:

Sets the processing and analysis target space.

<u>Current View</u>: The selected Z plane and the viewer area are processed. <u>Current Plane</u>: The selected Z plane is processed (XY).

<u>Current Time Point</u>: The selected time point is processed (XYZ).

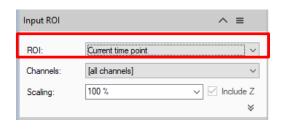
<u>Current Image Set</u>: The complete dataset (XYZ and time) is processed.

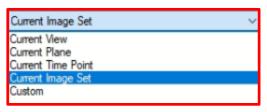
Custom: Allows to mix the previous methods.

Bounds: Sets the analysis area Bounds: edges. The whole XY bounds, the viewing area or a custom space can be applied.

<u>Planes</u>: Sets the analysis planes range. A single plane, a range of planes or the whole stack can be selected.

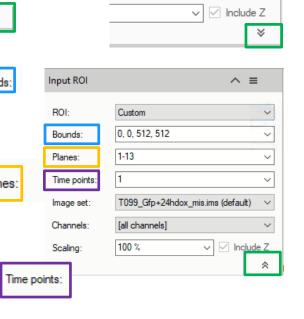
<u>Time Points</u>: Sets the analysis time points range. A single TP, a range of TPs or the whole movie can be selected.





DETAILS:

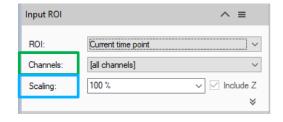
Use the Custom option during the pipeline setting and testing. Set a sub volume (XY, Planes, Time Points, channels) of your dataset on which perform the trial. This will speed up the setting process.



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Step 8.2.2 - Channels:

Sets the processing and analysis target channels. Selecting a single channel, all the operators in the pipeline will be forced to use it.



Step 8.2.3 – Scaling:

It scale the dataset reducing the size. The measurements will not be modified by the scaling factor.

TIPS:

Please refer to the (arivis Vision4D Help) for more details



Step 8.3

Change the Membrane based segmenter' operator parameters

Step 8.3.1 - Channels:

Sets the processing and analysis target channel(s).

Step 8.3.2

Min intensity: (membrane):

Set the Minimum membrane intensity (GL).

Step 8.3.3 - Split sensitivity:

Set the Split-Sensitivity coefficient.

Membrane based Segmenter

Preview:

Channel:

Ch3

Min intensity: (membrane):

→

Split sensitivity:

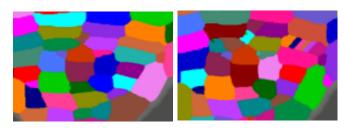
29 %

Max diameter: ✓

18,485 µm ✓

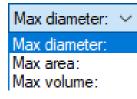
Max diameter: ✓

Decrising the Split sensitivity will fuse more objects.
Increasing the Split sensitivity will split more objects.



Step 8.3.4 – Max diameter:

Set the membrane maximum diameter (Area/Volume). Drop down the list to select the feature



TIPS:

Membrane Maximum Diameter (Area/Volume) can be measured directly from the dataset. Please refer to the (*arivis Vision4D Help*) for more details



Membrane based Segmenter

 $\wedge = \times$

Membrane based segmenter has the preview icon located on the panel header (title bar). Click on this icon to preview the operator result on the current plane / time point



Step 8.5

Add or remove operators from the pipeline

DETAILS:

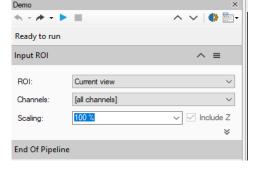
The Analysis Pipeline panel consists of two main areas. The Pipeline area and the analysis operations list area.

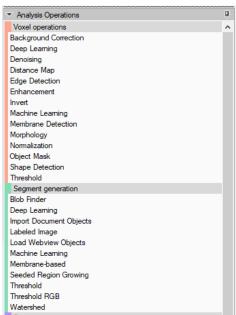
The *Operators* can be added to Pipeline in two ways

- 1. Double click on the *Operator* you wish to add to the current Pipeline. The *Operator* will be inserted at the end of the group of operations to which it belongs. Voxel Operations are positioned before the Segment generation meanwhile Store operations are put always at the end of the Pipeline.
- 2. Drag and drop the *Operator* you wish to add to the current Pipeline. The *Operator* will be automatically inserted in any place within the group of operations to which it belongs.

The *Operator* cannot be added during the Pipeline execution.

To remove an Operator from the Pipeline, press the X button located in the right side of the operator title bar.









TIPS:

Please refer to the (arivis Vision4D Help) for more details







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