

Application Note #14

How to create : «Freely XY Oriented Sub Volumes»

The application-note purpose is to guide the user in creating a sampling volume (ROI) freely oriented along X and Y axis. The application uses a Python script to create single or contiguous sub-regions that can be used as ROI for further analysis. The unique limits is related to the sampling volume shape, only regular 3D boxes are available.

Application Flowchart

Manual drawing of the reference ROI

- A 2D free hands ROI is drawn to define the orientation and the size of the sampling volume

Python Script (sub-regions creation)

- The Python script creates the contiguous sub-regions (sampling volume) freely oriented in the dataset volume

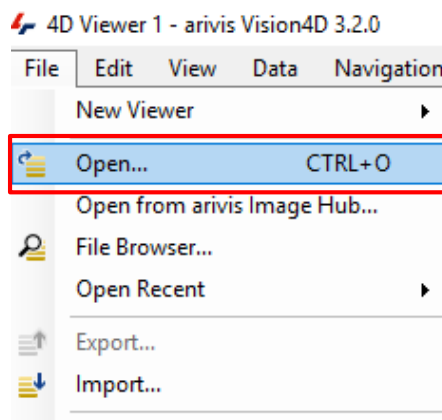
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1. Open the working dataset on Vision4D

Step 1.1

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

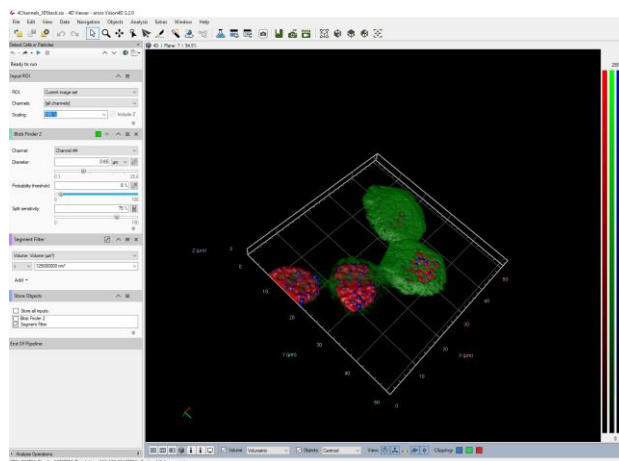


Step 1.2

Select the dataset from the file browser.

TIPS :

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



DETAILS:

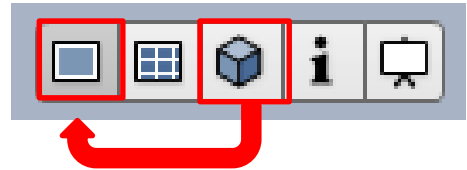
The dataset is a multi dimensional, discrete, representation of your real sample volume. It can be structured as a Z series of planes (Optical sectioning) of multiple channels (dyes) in a temporal sequence of time points (located in several spatial positions).

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)

2. Draw the reference ROI

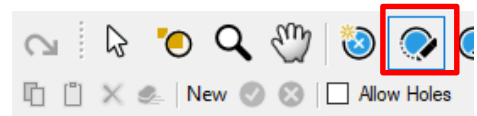
Step 2.1

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



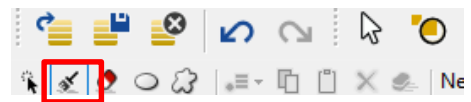
Step 2.2

Select the “Draw Objects Tool”



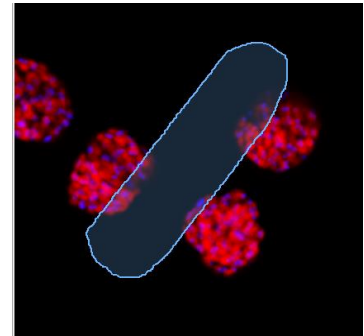
Step 2.3

Select the “Brush” tool



Step 2.4

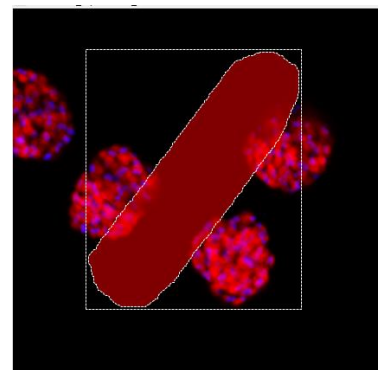
Draw the 2D ROI over any Z plane.
Use the «Erase Brush» to correct the ROI if necessary



Step 2.5

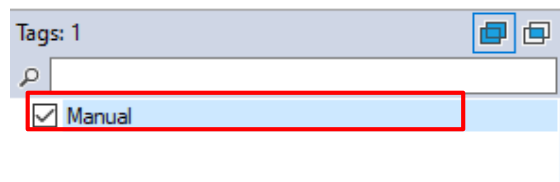


Press the green icon to confirm the ROI



DETAILS :

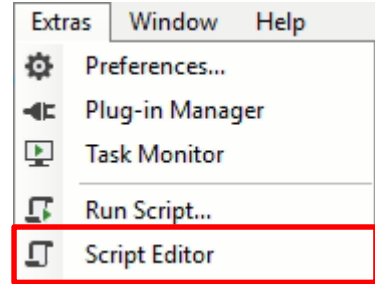
The TAG «Manual» is now available in the data table



3. Load the Python Script

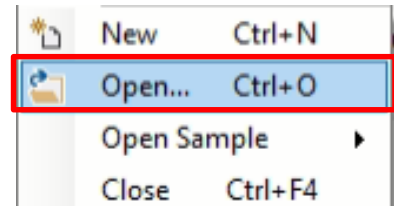
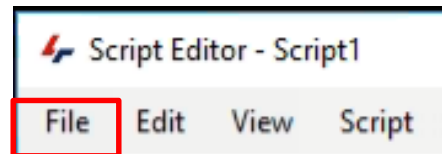
Step 3.1

Open Python Script Editor.
From the «*Extra*» menu, select the «*Script Editor*» item



Step 3.2

Load the “*Free-Oriented Sub-volume*” Python Script.
Browse the folder on which the file has been saved



Python script code overview

```
1 # coding: utf-8
2 # SCRIPT description
3 # CREATION DATA : 02/04/2019
4 # WRITTEN BY : Maurizio Abbate
5 # RELEASE 1.0.2
6 # RELEASE DATA : 21/05/2020
7 # PURPOSE : Create box segments freely orientated on XY
8 #
9 # NOTE :
10 #
11 # Tested for V4d Release : 3.1.4 / 3.2
12 #
13 # ----- External Package Import -----
14 import math as Math
15 import time
16 import aravis
17 import aravis_core as core
18 import aravis_objects as objects
19 # ----- End of external Package Import -----
20 #
21 # ##### USER SETTINGS #####
22 TAG_DESCRIPTOR = "Manual" #Cell Manual"
23 TAG_DESCRIPTOR_OUT = "Script" #Cell Manual"
24 COMPUTE_MAIN_BOX = False
25 FIRST_PLANE = 3 # -1 == bottom plane
26 LAST_PLANE = 10 # -1 == top plane
27 SIZE_IN_VOXELS = False
28 SIZE_BOX_HOR = 10.0 # used if SIZE_IN_VOXELS == True, expressed in um
29 SIZE_BOX_VER = 30.0 # used if SIZE_IN_VOXELS == True, expressed in um
30 NUM_BOX_LENGTH = 1 #NUM_BOX_VERT
31 NUM_BOX_WIDTH = 1 #NUM_BOX_HORIZ
32 NUM_BOX_DEPTH = 2 # Currently Not Used
33 # ##### END USER SETTINGS #####
```

NOTE :

Only the parameters located in the “*USER SETTING*” area can be modified.
Don't change any other number, definition or text in the code outside this dedicated area.

4. Set the Script features

In order to define the contiguous sub-regions (sampling volume) features, few parameters of the script should be adjusted to match your analysis needs. These parameters are located in the code area labeled as “**USER SETTING**”

```
# ##### USER SETTINGS #####
TAG_DESCRIPTOR = "Manual"          #Cell Manual"
TAG_DESCRIPTOR_OUT = "Script"      #Cell Manual"
COMPUTE_MAIN_BOX = False
FIRST_PLANE = 3                   # -1 == bottom plane
LAST_PLANE = 10                   # -1 == top plane
SIZE_IN_VOXELS = False
SIZE_BOX_HOR = 10.0               # used if SIZE_IN_VOXELS == True, expressed in um
SIZE_BOX_VER = 30.0               # used if SIZE_IN_VOXELS == True, expressed in um
NUM_BOX_LENGTH = 1                #NUM_BOX_VERT
NUM_BOX_WIDTH = 1                 #NUM_BOX_HORIZ
NUM_BOX_DEPTH = 2                 # Currently Not Used
# ##### END USER SETTINGS #####
```

Step 4.1

Set the Z planes range.

FIRST_PLANE defines the lower Z plane of the sub-regions ROI.

LAST_PLANE defines the higher Z plane of the sub-regions ROI.

The values of -1 set the Z planes range equal to the whole volume depth (total number of Z Planes available).

```
-----
FIRST_PLANE = -1
LAST_PLANE = -1
```

Step 4.2

COMPUTE_MAIN_BOX = True enables the creation of an additional ROI having the same sizes of the total sub-regions ROI size.

```
-----
COMPUTE_MAIN_BOX = False
```

Set the Script features (continue)

Step 4.3

NUM_BOX_LENGTH defines the number of sub-regions along the main axis (the longest one)

NUM_BOX_WIDTH defines the number of sub-regions along the minor axis (the shortest one).

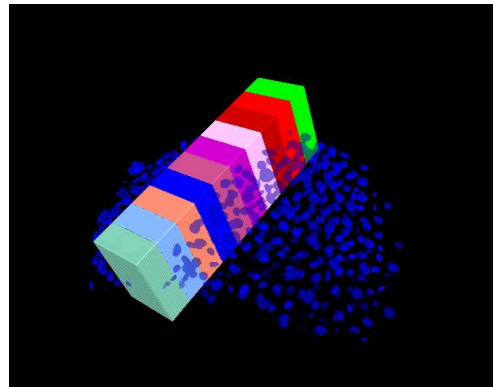
```
NUM_BOX_LENGTH = 14  
NUM_BOX_WIDTH = 2
```

TIPS :

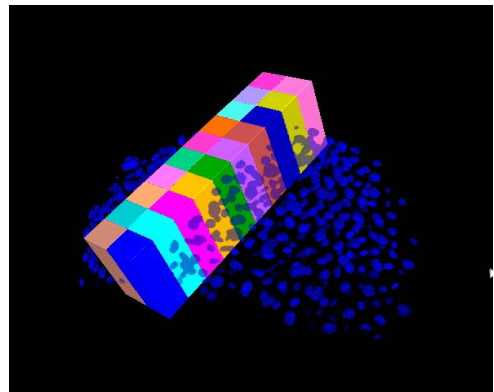
Set the number of sub-regions accordingly to the total size of the reference ROI . Don't create boxes too small.

Examples:

```
NUM_BOX_LENGTH = 10  
NUM_BOX_WIDTH = 1
```



```
NUM_BOX_LENGTH = 10  
NUM_BOX_WIDTH = 2
```



Set the Script features (continue)

Step 4.4

SIZE_IN_VOXELS defines if the size of the Sub-Volume is expressed in metric unit (True) or it is calculated from the reference ROI size (False).

```
SIZE_IN_VOXELS = False  
SIZE_BOX_HOR = 10.0  
SIZE_BOX_VER = 30.0
```

Step 4.5

SIZE_BOX_HOR and SIZE_BOX_VER defines the Sub-Volume XY size in microns. *The sizes are referred to a single box.*

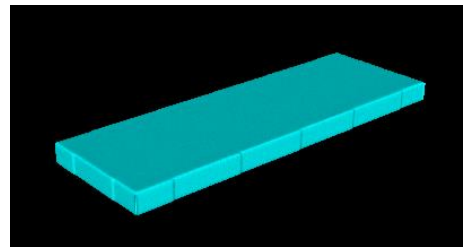
SIZE_IN_VOXELS must be **True** to create the box in microns.

NOTE :

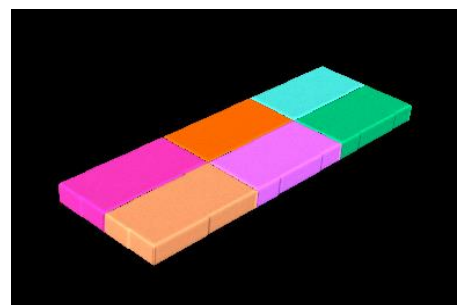
NUM_BOX_LENGTH and NUM_BOX_WIDTH are also involved in the metric Sub-Volume creation.

Examples:

```
SIZE_IN_VOXELS = True  
NUM_BOX_LENGTH = 1  
NUM_BOX_WIDTH = 1  
SIZE_BOX_HOR = 10 (microns)  
SIZE_BOX_VER = 30 (microns)
```



```
SIZE_IN_VOXELS = True  
NUM_BOX_LENGTH = 3  
NUM_BOX_WIDTH = 2  
SIZE_BOX_HOR = 5 (microns)  
SIZE_BOX_VER = 10 (microns)
```



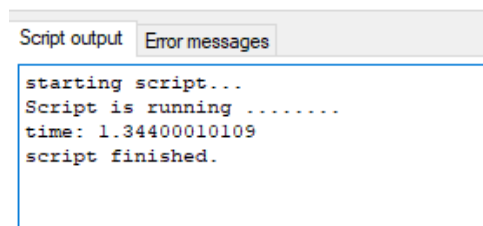
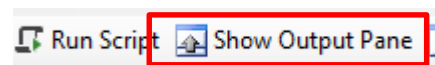
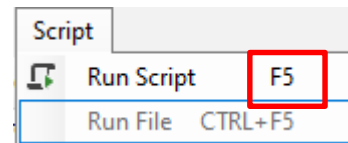
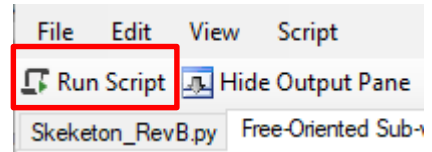
5. Run the Python Script

Step 5.1

Run the “*Free-Oriented Sub-volume*” Python Script pressing the “Run Script” button or pressing the F5 key.

TIPS :

Activate, if not already displayed, the “Output Panel”. The status of the script execution (errors including) will be visualized here





Contact the arivis local area sales manager to get more information about how to get the python script mentioned here.

Contact the arivis application support to receive additional technical details about the topic described in the application note, or how to adapt the application workflow to your requirements.

“The quantitative analysis of the images represents the art of transforming a visual sensation into its schematic and discrete form allowing its univocal description, classification and mathematical and logical interpretation of its spatial and temporal components”

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