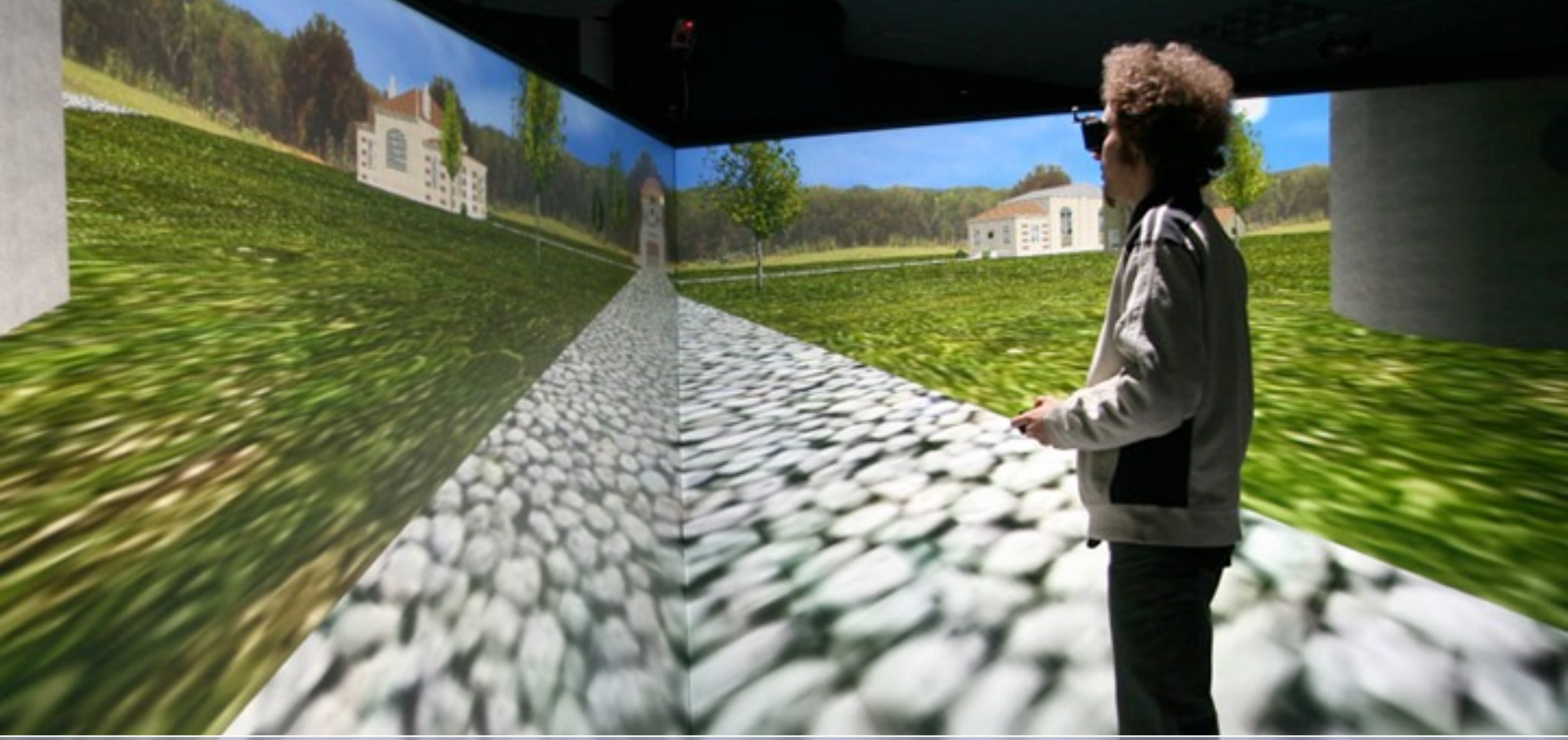
Greetings from Hanchuan
wacom

“工欲善其事，必先利其器”

-《论语·魏灵公》，~470 BC

“Sharpen the knife before cutting the wood”

-《Analects of Confucius》，~470 BC, China

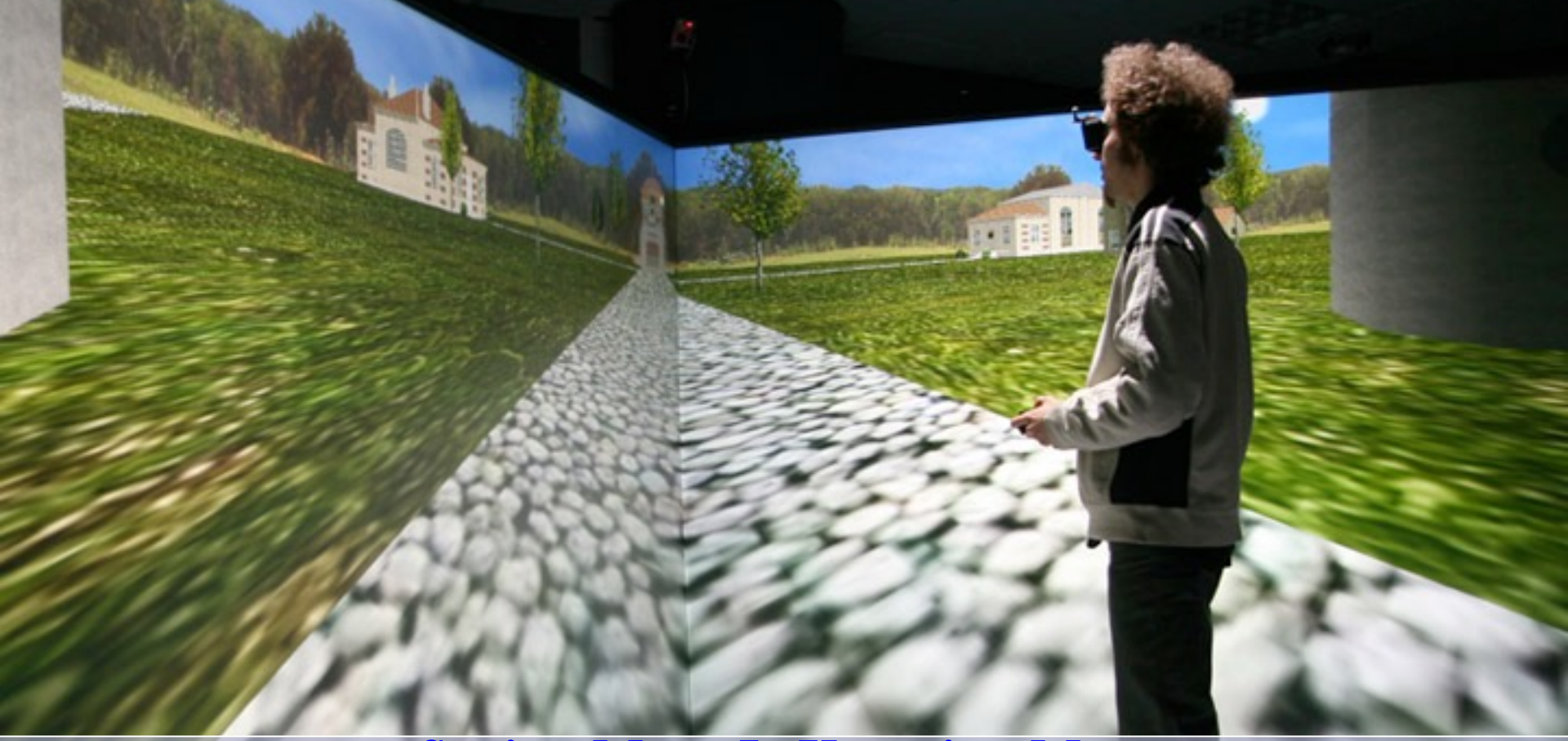


Seeing More Is Knowing More
- How Vaa3D Can Help Biology

Hanchuan Peng

a talk in 2011

Picture from:
http://www.antycipsimulation.com/solutions/virtual-reality-solutions



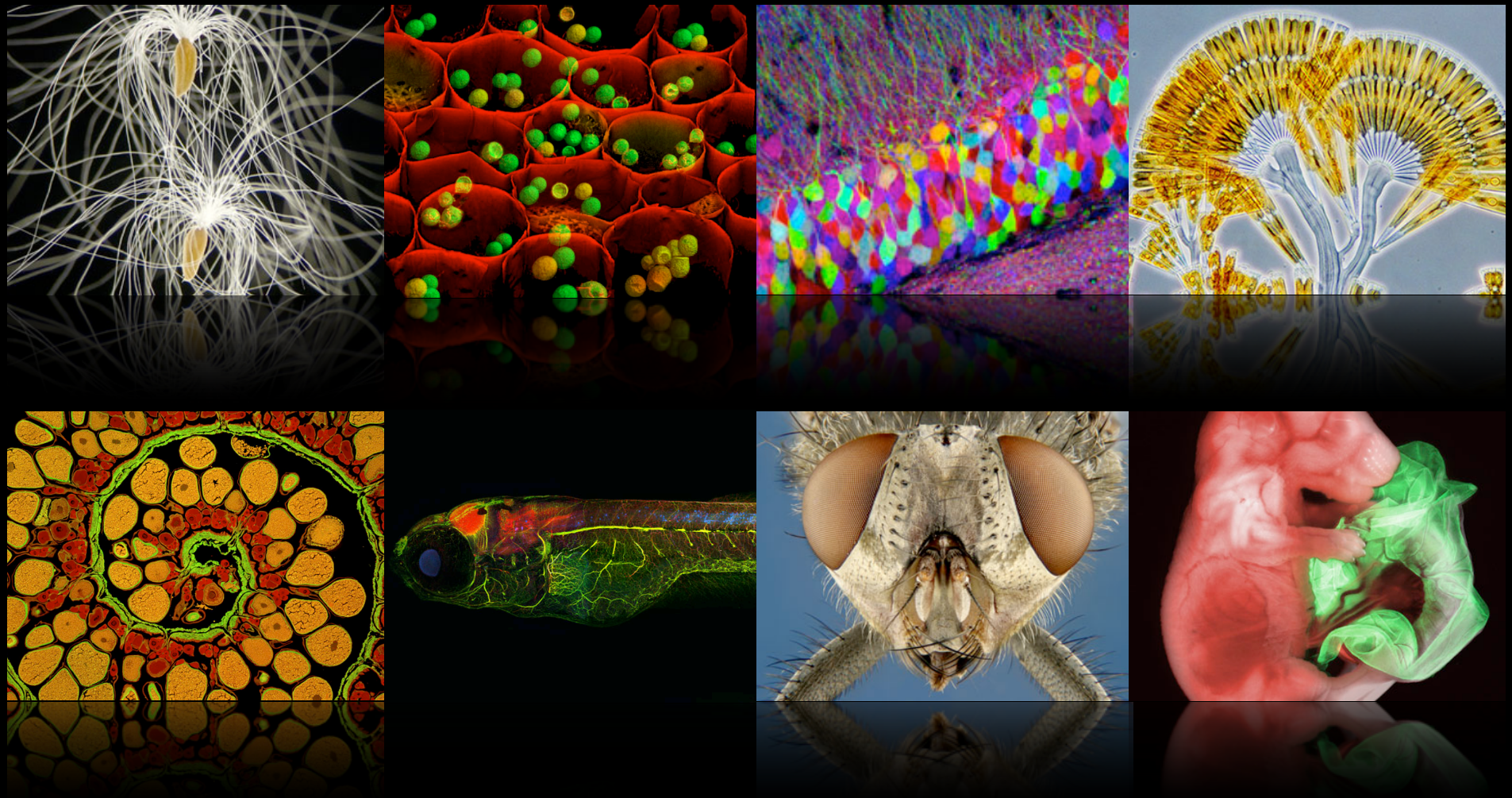
~~Seeing More Is Knowing More~~
~~- How Vaa3D Can Help Biology~~

Evolving Vaa3D: The Best is Yet to Come

Hanchuan Peng

Picture from:
http://www.antycipsimulation.com/solutions/virtual-reality-solutions

Bioimages form a major type of data for computational biologists & bioinformaticians



Pictures: courtesy of Nikon Small World of Competition

There are several

significant 3D image computing challenges…

besides

the usually called “image analysis”

3D Visualization problem.

How to see large (≥gigabytes, e.g. 1024x1024x512xRGB=1.5G) 3D/4D/5D images, on $1,000~$2,000 PCs using real-time 3D rendering (e.g. MIP)?

3D Interaction & Prior Generation problem.

How to conveniently incorporate a user' input (e.g. 3D point/curve/region of interest) in image stacks directly in 3D to guide image analysis?

3D Proof-editing problem.

How to conveniently (e.g. drag & drop) overlay any surface objects (produced by image processing, e.g. segmented cells, traced neurons, segmented brain regions) with large image stacks directly in 3D, and do proof-reading and editing in 3D right away?

nature biotechnology

V3D enables real-time 3D visualization and quantitative analysis of large-scale biological image data sets

Hanchuan Peng, Zongcai Ruan, Fuhui Long, Julie H Simpson & Eugene W Myers

The V3D system provides three-dimensional (3D) visualization of gigabyte-sized microscopy image stacks in real time on current laptops and desktops. V3D streamlines the online analysis, measurement and proofreading of complicated image patterns by combining ergonomic functions for selecting a location in an image directly in 3D space and for displaying biological measurements, such as from fluorescent probes, using the overlaid surface objects. V3D runs on all major computer platforms and can be enhanced by software plug-ins to address specific biological problems. To demonstrate this extensibility, we built a V3D-based application, V3D-Neuron, to reconstruct complex 3D neuronal structures from high-resolution brain images. V3D-Neuron can precisely digitize the morphology of a single neuron in a fruitfly brain in minutes, with about a 17-fold improvement in reliability and tenfold savings in time compared with other neuron reconstruction tools. Using V3D-Neuron, we demonstrate the feasibility of building a 3D digital atlas of neurite tracts in the fruitfly brain.

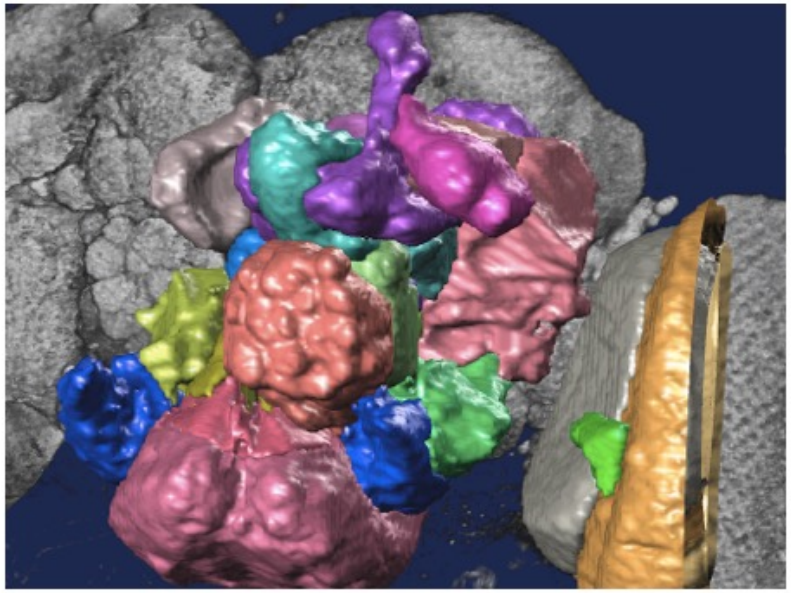
Janelia Farm Research Campus, Howard Hughes Medical Institute, Ashburn, Virginia, USA. Correspondence should be addressed to H.P. (pengh@janelia.hhmi.org).

Received 30 November 2009; accepted 8 February 2010; published online 14 March 2010; doi:10.1038/nbt.1612

VAA3D 挖三维

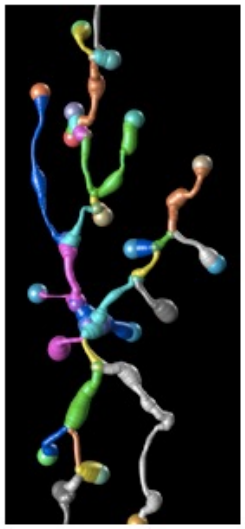
ABOUT VAA3D | MAJOR FUNCTIONS | DOWNLOAD | TOOLBOXES AND PLUGINS | HOW TO USE
TUTORIAL MOVIES | TEST DATA | PUBLICATIONS | LICENSE | CONTACT | HACKATHON

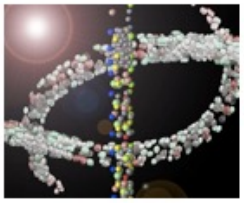
Vaa3D lets you explore & understand your 3D/4D/5D images a lot easier!

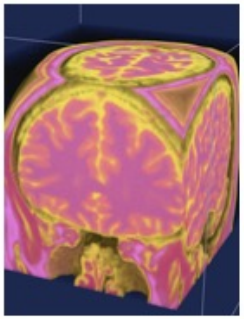


Vaa3D: A Swiss army knife for bioimage visualization & analysis

Vaa3D [1, 2, 3] is a handy, fast, and versatile 3D/4D/5D Image Visualization & Analysis System for Bioimages & Surface Objects. It also provides many unique functions ... It is also Open Source, supports a very simple and powerful plugin interface and thus can be extended & enhanced easily...







Real-time 3D visualization of large (gigabyte sized) image stacks, and 3D WYSIWYG functions, thus efficient ways for real-time, quantitative measurement & analyses (e.g. neuron tracing).

Peng, et al,
***Nature Biotechnology*, 2010.**
Nature Communications, 2014, 2019; Nature Methods, 2009/2012a/2012b/2016/2017 …

Open Source & Free software:
http://vaa3d.org

Significant use in others' work: Science, Cell, Nature Methods, Nature Neurosci, Cytometry, IEEE TMI, Bioinfo, MICCAI, and many others.

Since we don’t want to reinvent the same wheel

(or even its components)

again & again & again

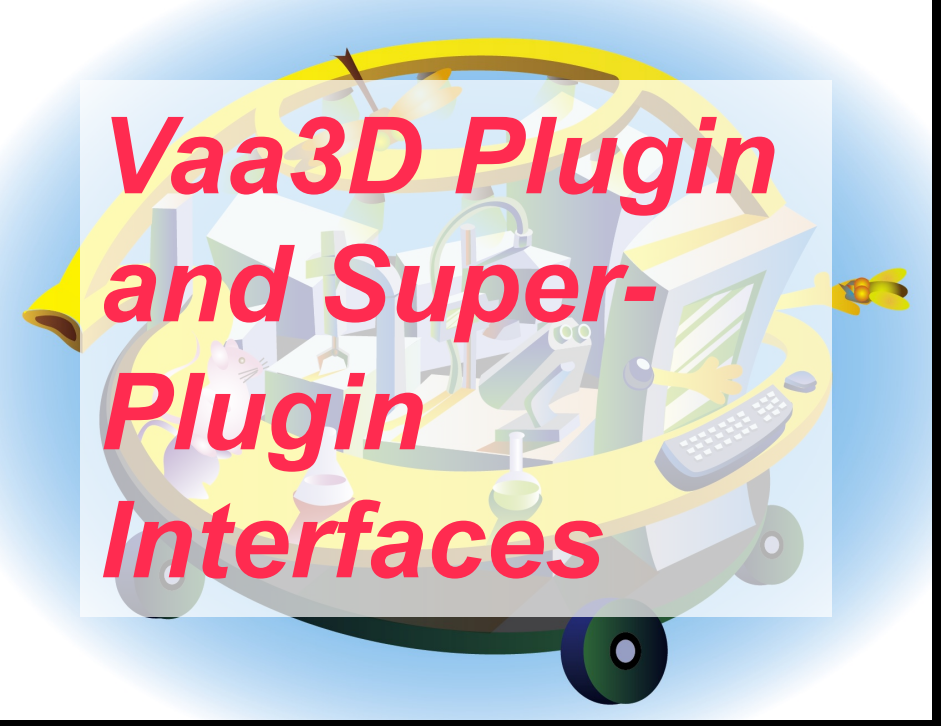
for 3D image computing problems of different natures,

it becomes interesting to consider

how to “scale-up” a finite number of modules

to solve a combinatorial number of image computing problems.

Scale-up the Applicability of Bioimage Informatics Tools Using Vaa3D Plugin and Super-Plugin Interfaces



BioImage analysis modules

Online help & real-time usage listing; deploy a plugin at run-time, without reloading image data, …

Bioimaging (3D/4D/5D image acquisition, microsurgery, …)

3D/4D surface data computing

Bioimage databases

System event (mouse, keyboard, etc) filtering

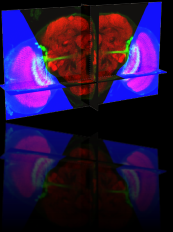
Pipelining different modules (plugin-call-plugin, real-time DLL hand-shaking, command-line binding)

Computing many/arbitrary-number images/surfaces at the same time

Invoke external tools (ITK, Java, R, Matlab, …)

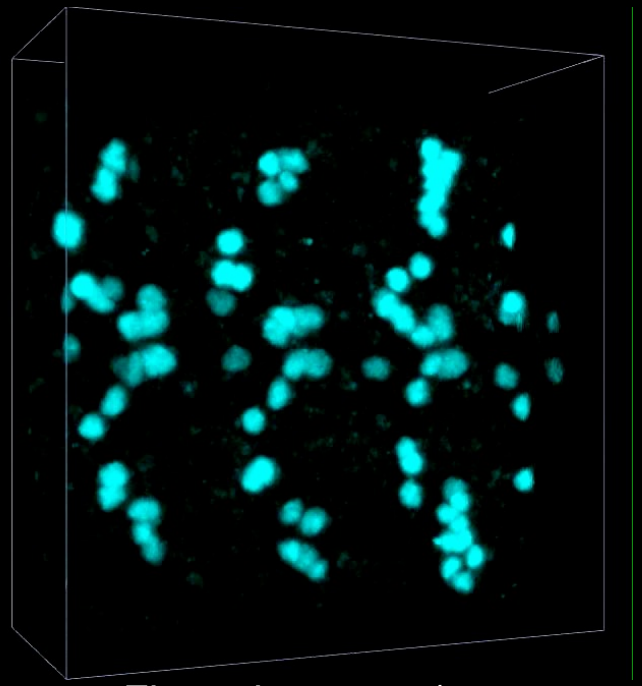
Pluginize everything and make the program much more modulized and reconfigurable

Invoked by external tools (web apps, Matlab, …)



Vaa3D contains a number of 3D image/pattern analysis & management plugins

- **Segmentation**
  - globular objects (cell bodies, nuclei, bouton)
  - fibrous objects (neurons)
  - Irregular shaped (brain compartments)
- **Registration**
  - global (affine, rigid), local (non-rigid, elastic)
  - Feature point detection, matching
  - Generate of warping field
  - Cutting plane restacking along curve
- **Classification**
  - Image [region] classification
  - Neuron structure comparison & categorization
- **Data management**
  - 3D landmarking
  - Proof-reading
  - Manage images and associated meta data
  - Manage large image archive, e.g. aligned images & atlas files
- **Almost 300 user-developed plugins (>160 released, many other under the review folder, - source code available)**

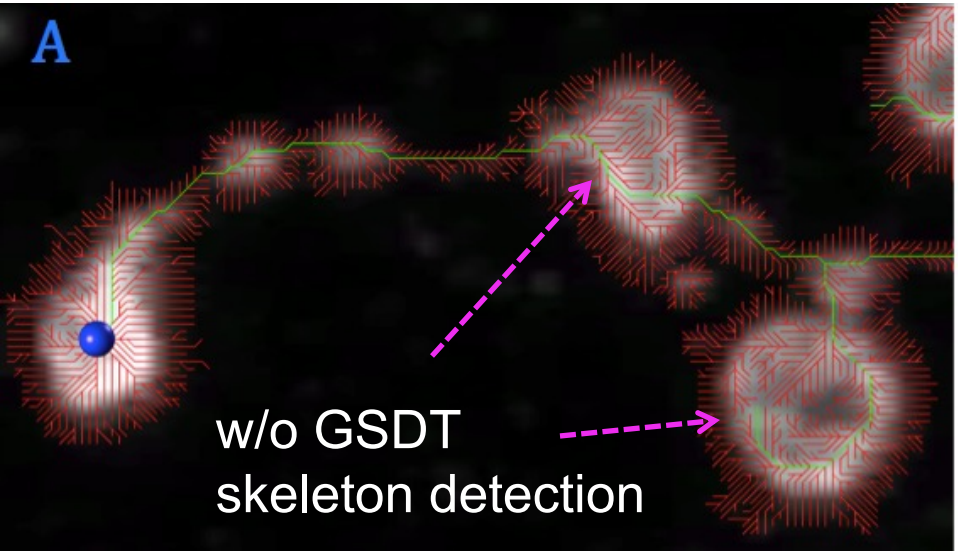


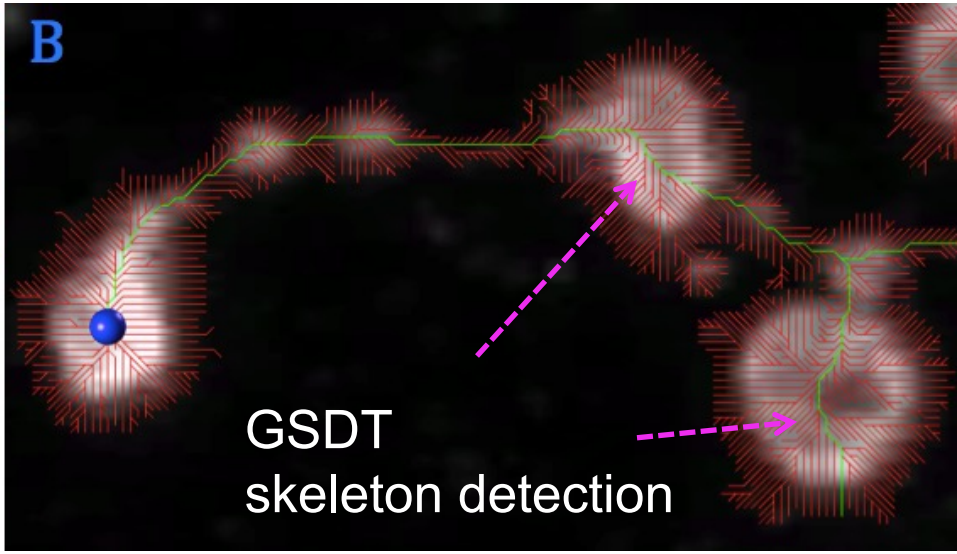
Fly-embryo seg (see Fuhui Long talk)

EM Image Seg: http://www.youtube.com/watch?v=haO3WeYq37E&feature=player_embedded

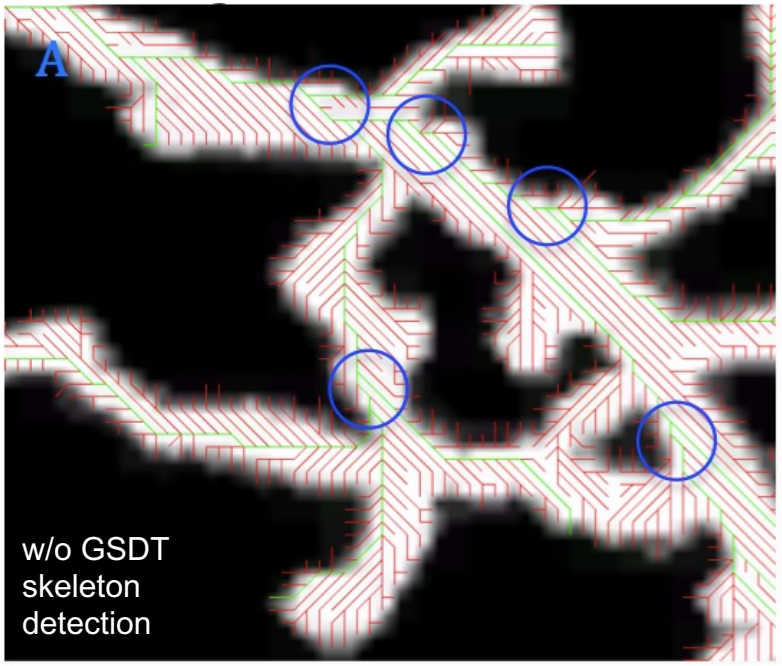
Some 3D image computing plugins

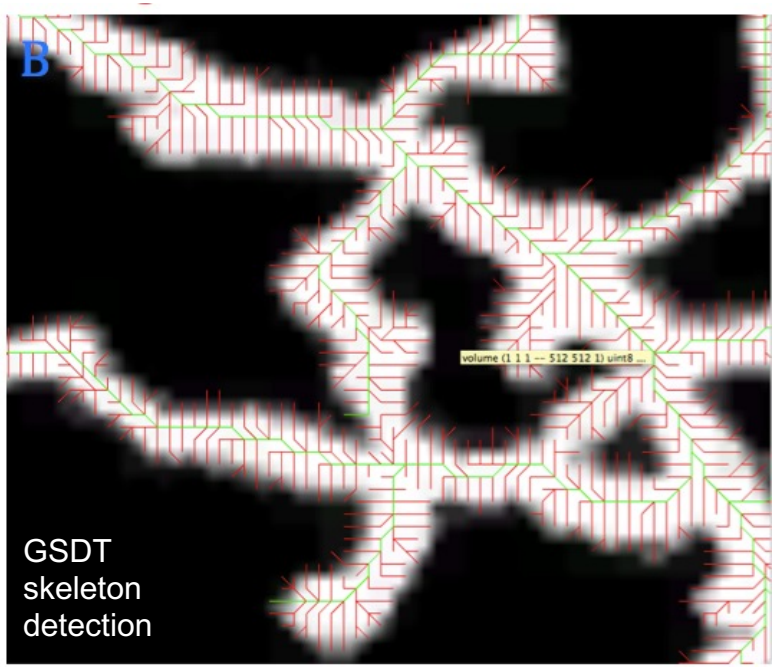
GSDT: GrayScale Distance Transform



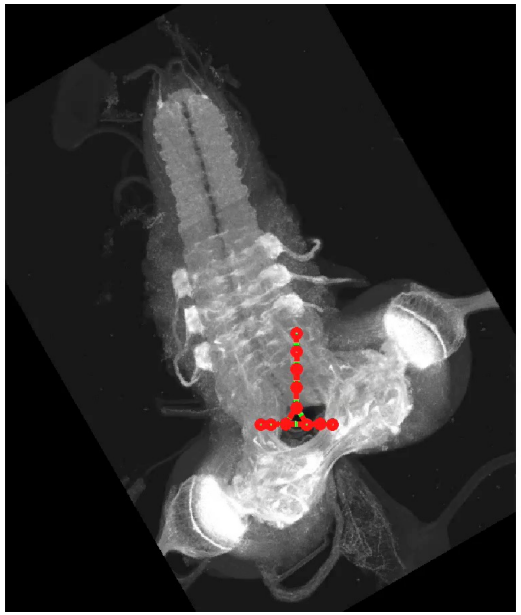


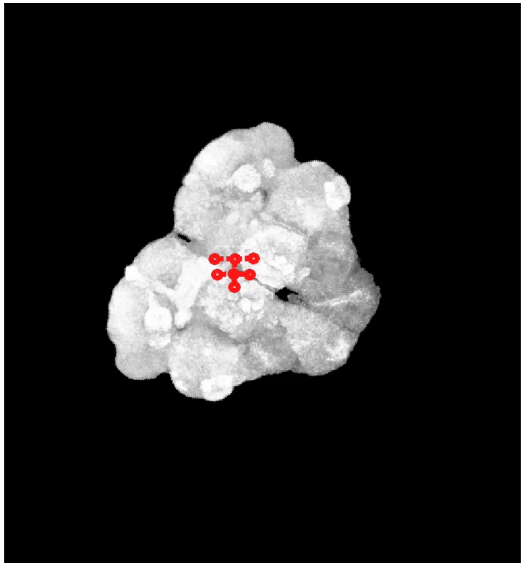
Xiao & Peng 2012





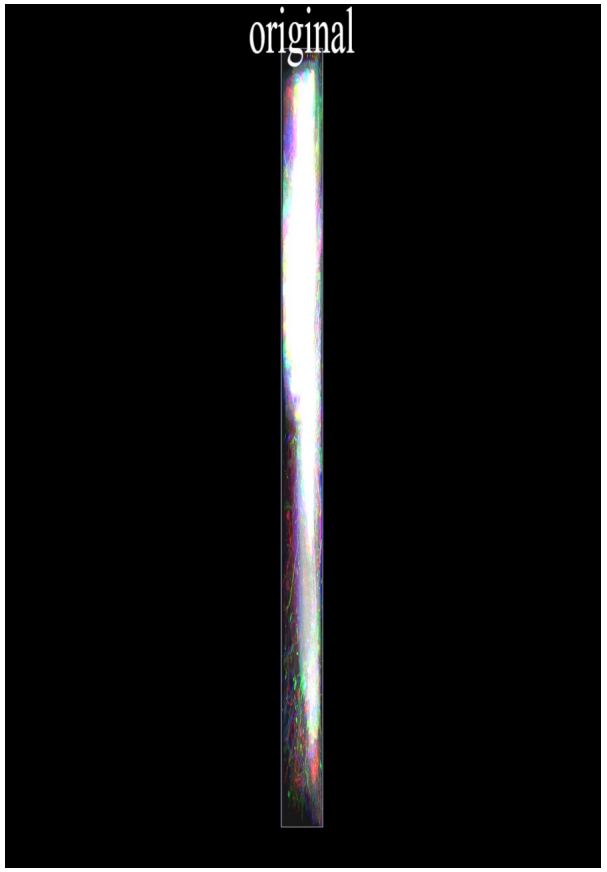
Principal Skeleton Deformable Model & Its Use in Image Alignment





Vaa3D-Principal Skeleton plugins.

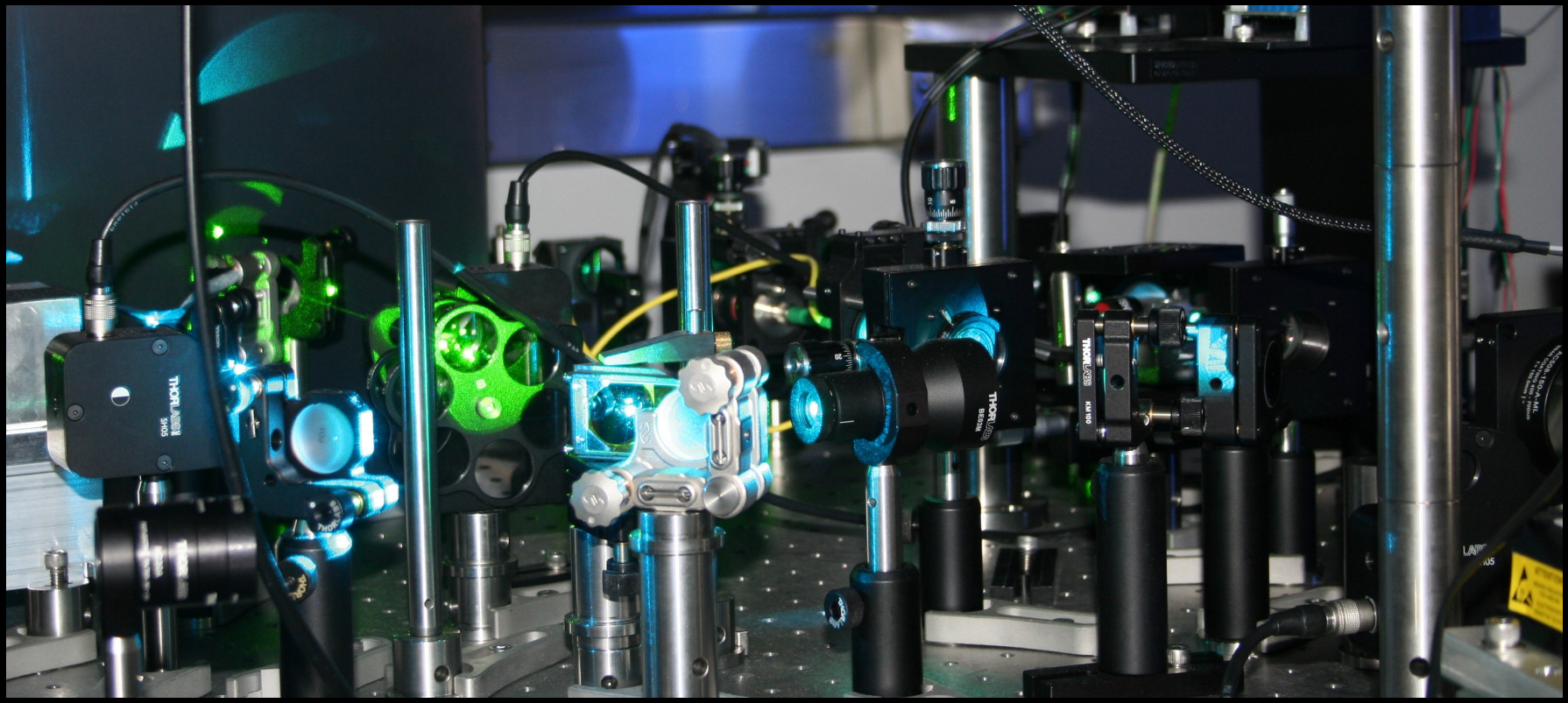
Qu & Peng, Bioinformatics, 2010.



237 overlaid & color-coded larval images

3D imaging & microsurgery plugins

Hanchuan Peng's SmartScope: Prototype I (Sept-Oct 2010).



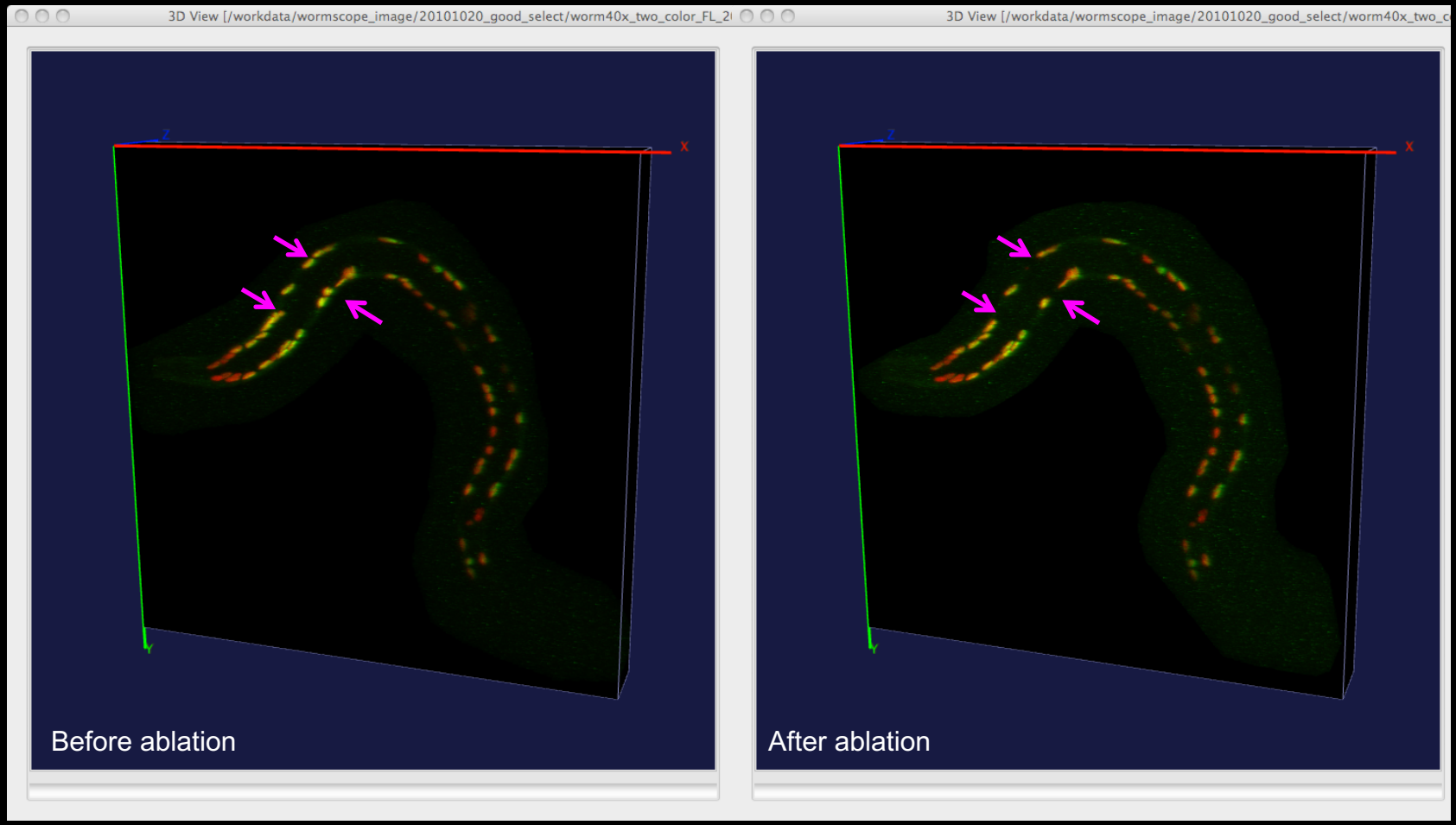
detectors here (oov)

objective & camera here (oov)

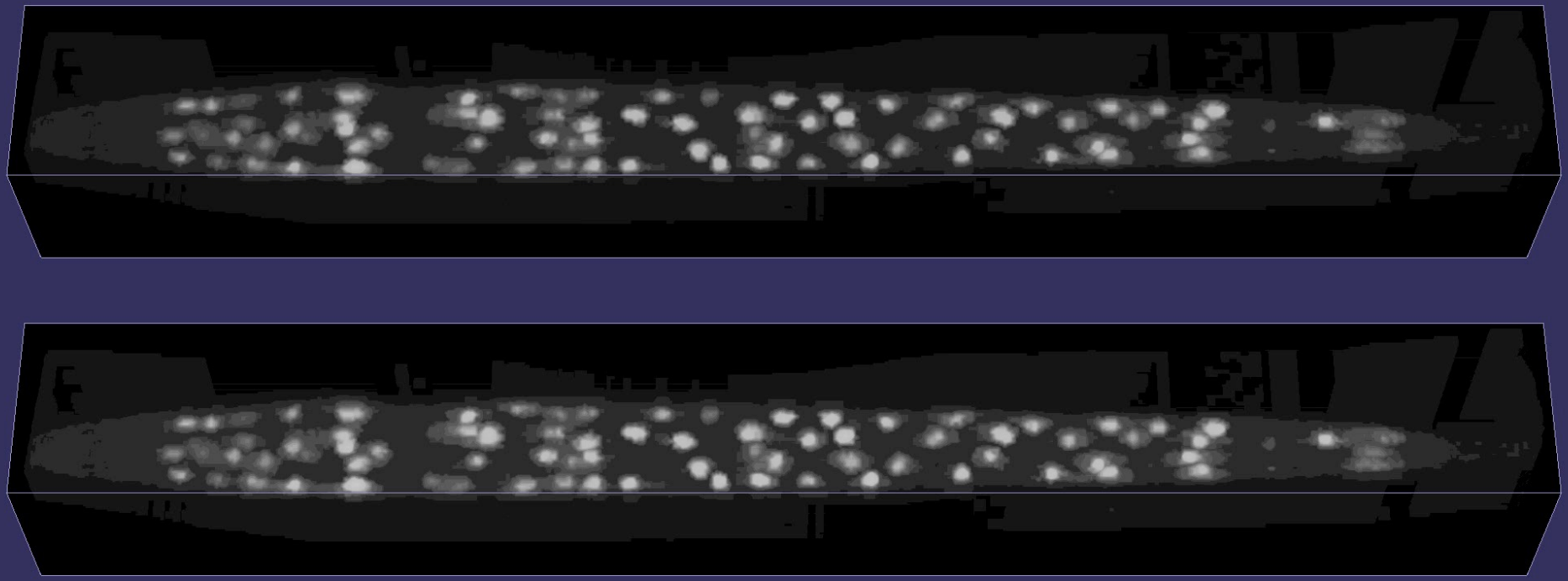
ablation laser here (oov)

Other out-of-view (OOV) parts:
Shutters, amplifiers, controlling computer, etc.

3D instant cell-ablation/bleaching and potential optogenetic experiments



This Vaa3D-SmartImaging plugin combines with another Vaa3D-SRS plugin for automated cell-targeting (stimulation / recording) in 3D, for the goal to decode the functional wiring map of a worm's brain

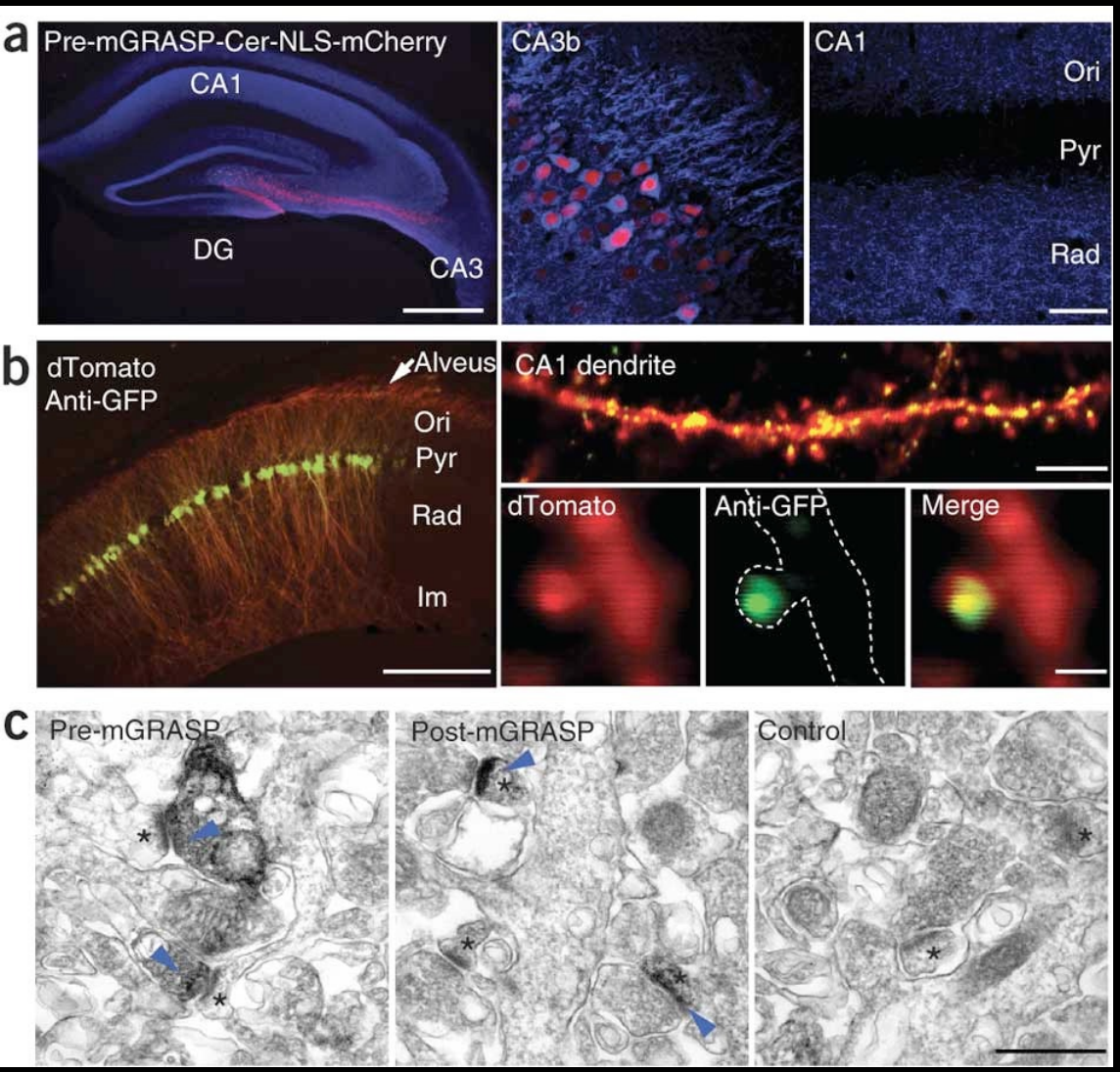


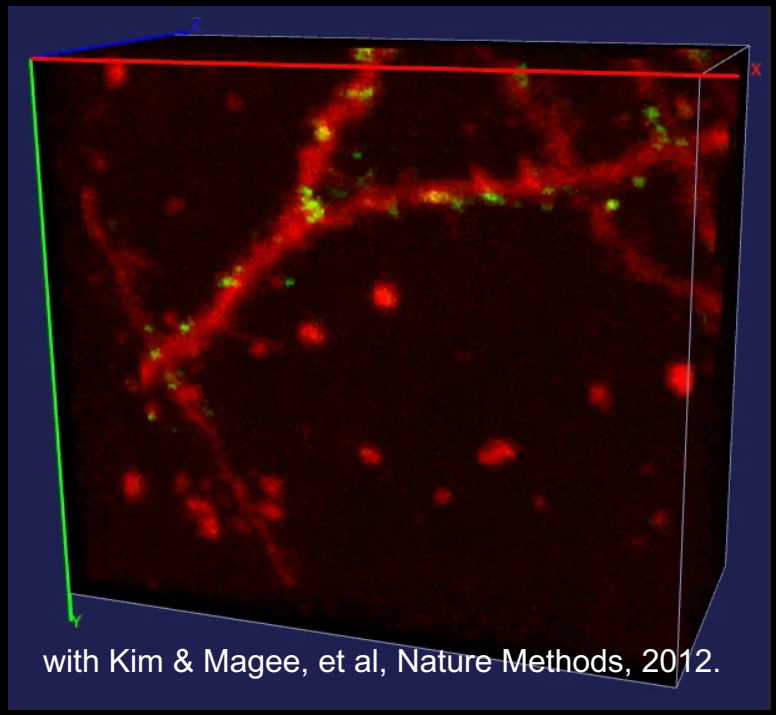
$$E(P)=\sum_{j=1}^{K}\sum_{i=1}^{N} p_{ij}\,\Psi(v_i)\,D\left(\theta(\overrightarrow{v_i c_j},\overrightarrow{\mathrm{AP}}),\gamma\right)\frac{\|v_i-c_j\|^2}{Q^2}-T\cdot H(P) \qquad \Psi(v_i)=255-I(v_i)$$

$$c_j^{\mathrm{new}}=f(u_j,C^0) \quad \text{where} \quad u_j=\sum_{i=1}^{N} p_{ij}v_i. \qquad f(u_j,C^0)=f(u_j,\{c_1^0,c_2^0,\ldots,c_K^0\})$$

$$=A\times c_j^0+\sum_{k=1}^{K}\omega_k\cdot\phi(\|u_j-c_k^0\|)$$

This Vaa3D-SmartImaging plugin may also combine with another Vaa3D-synapse-detection plugin for ablating individual synapses





(a) Distribution of Cre recombinase–independent pre-mGRASP (aavCAG–pre-mGRASP–mCerulean-2A-nls-mCherry; pre-mGRASP-Cer-NLS-mCherry) visualized with blue fluorescence signals from mCerulean. Overview of the hippocampus (left) and high-magnification images of subregions (CA3b and CA1) show infected cell nuclei in red (mCherry) and their axonal projections in blue (pre-mGRASP–mCerulean). Ori, stratum oriens; pyr, stratum pyramidale; and rad, stratum radiatum. Scale bars, 500 μm (left) and 40 μm (right). (b) Dendritic distribution of post-mGRASP (paavCAG-post-mGRASP-2A-dTomato) in CA1 pyramidal neurons visualized by fluorescence immunostaining using polyclonal antibody to GFP (anti-GFP). Cytosolic signal from dTomato is indicated with an arrow (left). High-magnification images show that post-mGRASP appears to be enriched in the postsynaptic density (right). Lm, stratum lacunosum-moleculare. Scale bars, 250 μm (left), 5 μm (right top) and 1 μm (right bottom). (c) Immunoelectron microscopy images of pre-mGRASP, post-mGRASP and nontransduced hippocampi as a control using polyclonal anti-GFP. Asterisks indicate postsynaptic density, and blue arrowheads indicate immuno-silver-gold particles. Scale bar, 500 nm.

Auto-detect Mouse-GRASP labeled synapses (based on Vaa3D)

Pipelining many plugins using a super-plugin

Listing of ~170 Released Vaa3D Plugins (v2.801)

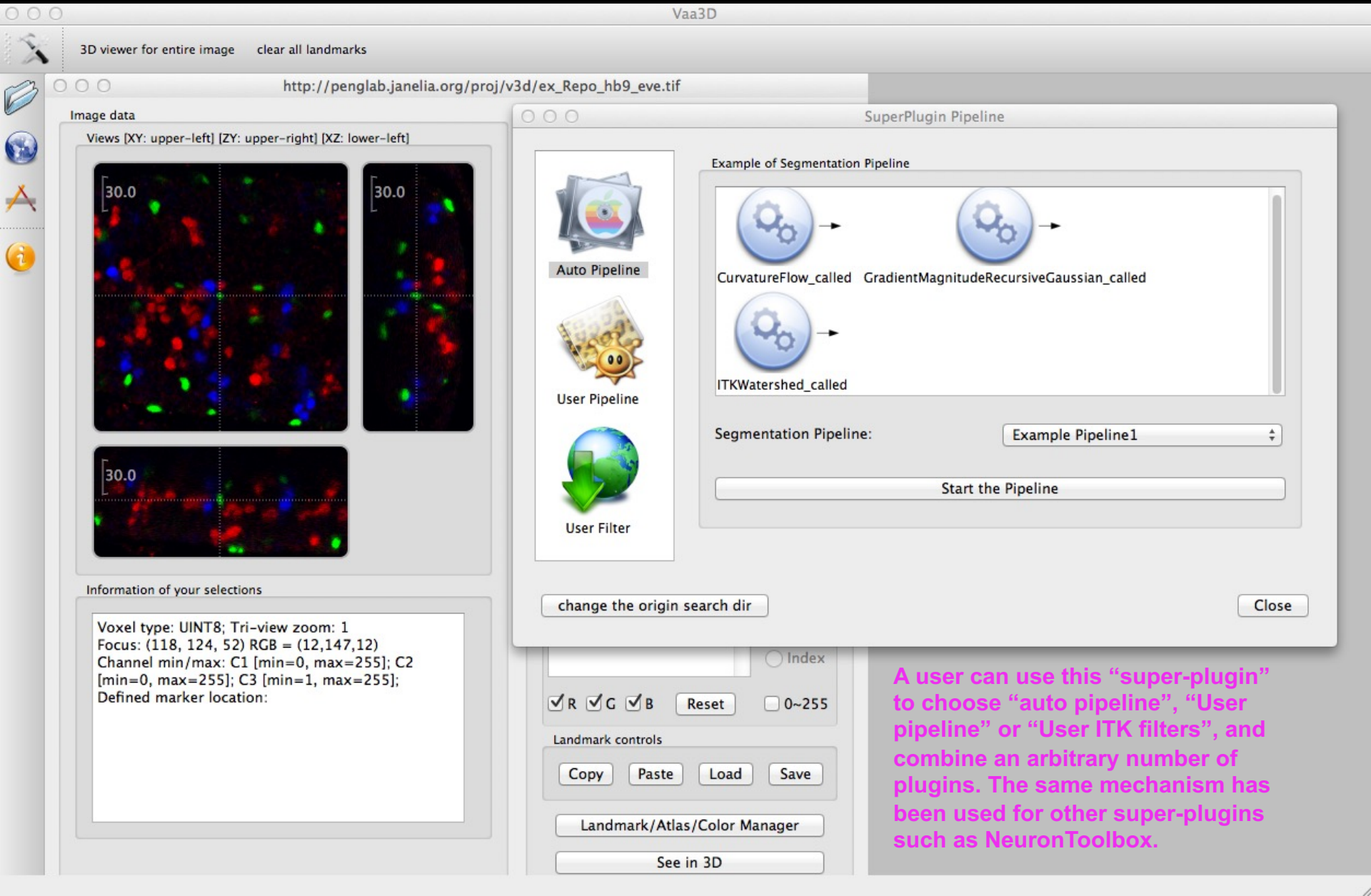
```
pengh-lm6:work pengh$ v3d -h

Vaa3D: a 3D image visualization and analysis platform developed by Hanchuan Peng and colleagues.

Usage: v3d -h -M moduleCode [all other options specific to different modules]
    -h/H         help information.
    -i <file>                    open single or multiple image (.tif/.tiff, .lsm, .mrc, .raw/.v3draw) / object (.ano, .apo, .swc, .marker) files
    -o <file>                    indicates single or multiple outputs
    -x <plugin_dll_full_path or unique partial path>    a string indicates the full path or a unique partial path of a dll (for a plugin) to be launched.
    -m <menu_name>               a string indicates which menu of a plugin will be called.
    -f <function_name>           a string indicates which function of a plugin will be called.
    -p <parameters>              a string indicates parameters that plugin function use
    -pf <configuration>          a string read from configuration file indicates parameters that plugin function use
    -v                           force to open a 3d viewer when loading an image, otherwise use the default v3d global setting (from "Adjust Preference")
    -na                          open NeuronAnnotator work-mode directly
    -cmd  [headless command-line arguments, intended for compute grid use. Try '-cmd -h' for more information on this option]
Searching in ./plugins ......
Searching ./plugins done.

Found [168] plugins
#1          /Users/pengh/work/v3d_external/bin/plugins/_Vaa3D_plugin_creator/libplugin_creator_debug.dylib
#2          /Users/pengh/work/v3d_external/bin/plugins/celegans/atlasguided_seganno/libplugin_atlasguided_stranno_partial_debug.dylib
#3          /Users/pengh/work/v3d_external/bin/plugins/celegans/celegans_straighten/libcelegans_straighten_debug.dylib
#4          /Users/pengh/work/v3d_external/bin/plugins/color_channel/split_extract_channels/libchannelsplit_debug.dylib
#5          /Users/pengh/work/v3d_external/bin/plugins/data_IO/load_image_using_Bioformats/libimageIO_Bioformat_debug.dylib
#6          /Users/pengh/work/v3d_external/bin/plugins/data_type/5D_Stack_Converter/libmovieZCswitch_debug.dylib
#7          /Users/pengh/work/v3d_external/bin/plugins/data_type/Convert_8_16_32_bits_data/libdatatypeconvert_debug.dylib
#8          /Users/pengh/work/v3d_external/bin/plugins/data_type/Convert_Image_to_AtlasViewMode/libconvert_img_to_atlasview_debug.dylib
#9          /Users/pengh/work/v3d_external/bin/plugins/data_type/intensity_rescale/librescale_debug.dylib
#10          /Users/pengh/work/v3d_external/bin/plugins/histogram/libhistogram_debug.dylib
#11          /Users/pengh/work/v3d_external/bin/plugins/hp/libhp_debug.dylib
#12          /Users/pengh/work/v3d_external/bin/plugins/image_blending/blend_multiscanstacks/libblend_multiscanstacks_debug.dylib
#13          /Users/pengh/work/v3d_external/bin/plugins/image_edge_detection/Edge_Extraction_from_Mask_Image/libedge_of_maskimg_debug.dylib
#14          /Users/pengh/work/v3d_external/bin/plugins/image_filters/Fast_Distance_Transform/libdt_debug.dylib
#15          /Users/pengh/work/v3d_external/bin/plugins/image_filters/Gaussian_Filter/libgaussianfilter_debug.dylib
#16          /Users/pengh/work/v3d_external/bin/plugins/image_filters/Grayscale_Image_Distance_Transform/libgsdt_debug.dylib
#17          /Users/pengh/work/v3d_external/bin/plugins/image_filters/Laplacian_Filter/liblaplacianfilter_debug.dylib
#18          /Users/pengh/work/v3d_external/bin/plugins/image_filters/min_Max_Filter/libminMaxfilter_debug.dylib
#19          /Users/pengh/work/v3d_external/bin/plugins/image_geometry/Montage_All_Z_Sections/libmontage_image_sections_debug.dylib
#20          /Users/pengh/work/v3d_external/bin/plugins/image_geometry/ReCenter_Image/librecenterimage_debug.dylib
#21          /Users/pengh/work/v3d_external/bin/plugins/image_geometry/Rotate_Image/librotateimg90_debug.dylib
#22          /Users/pengh/work/v3d_external/bin/plugins/image_registration/littleQuickWarp/liblittlequickwarp_debug.dylib
#23          /Users/pengh/work/v3d_external/bin/plugins/image_registration/SSD_registration/libplugin_PQ_imagereg_debug.dylib
#24          /Users/pengh/work/v3d_external/bin/plugins/image_resolution/XYZ_Resolution/libexample_reset_xyz_resolution_debug.dylib
#25          /Users/pengh/work/v3d_external/bin/plugins/image_ROI/ROI_Editor/libroi_editor_debug.dylib
#26          /Users/pengh/work/v3d_external/bin/plugins/image_segmentation/Label_Objects/libregiongrow_debug.dylib
#27          /Users/pengh/work/v3d_external/bin/plugins/image_segmentation/lobeseger/liblobeseg_debug.dylib
```

Vaa3D-ITK Pipelining GUI



A user can use this "super-plugin" to choose "auto pipeline", "User pipeline" or "User ITK filters", and combine an arbitrary number of plugins. The same mechanism has been used for other super-plugins such as NeuronToolbox.

A Vaa3D Plugin contains essentially only FOUR simple interfacing functions

```
#ifndef __EXAMPLE_PLUGIN_H__
#define __EXAMPLE_PLUGIN_H__

#include <QtGui>
#include <v3d_interface.h>

class ExamplePlugin : public QObject, public V3DPluginInterface2_1
{
        Q_OBJECT
        Q_INTERFACES(V3DPluginInterface2_1);

public:
        float getPluginVersion() const {return 1.1f;}

        QStringList menulist() const;
        void domenu(const QString &menu_name,
                    V3DPluginCallback2 &callback,
                    QWidget *parent);

        QStringList funclist() const ;
        bool dofunc(const QString &func_name,
                    const V3DPluginArgList &input,
                    V3DPluginArgList &output,
                    V3DPluginCallback2 &callback,
                    QWidget *parent);
};

#endif
```

Menu items in GUI

The actual action(s) of each menu item

Function items for any other purposes

The actual action(s) of each function

Vaa3DPlugins – vaa3d – Introduction of Vaa3D plugins – A Swiss army knife for bioimage visualization & analysis – Google Project Hosting

Vaa3DPlugins – vaa3d – Introd...

code.google.com/p/vaa3d/wiki/Vaa3DPlugins

vaa3d plugin

My favorites ▾ | Sign in

vaa3d

A Swiss army knife for bioimage visualization & analysis

Search projects

Project Home | **Wiki**

Search Current pages for Search

~~GoogleCode~~

GitHub for Vaa3D

Plugins

- How to Use Vaa3D
- Plugins
  - Overview
  - Plugins list
  - Command line access
  - Write a plugin: example
  - Compile plugins
  - Plugin-API details
- Toolboxes
  - Matlab I/O
  - Matlab bind
  - ImageJ/Fiji
  - Bioformats
- Under the Hood
- About
- Other links

Vaa3DPlugins

Introduction of Vaa3D plugins

Featured

Content

- What are Vaa3D plugins?
  - Where to find Vaa3D plugins' executable?
  - Where to find Vaa3D plugins' source code? How to batch-compile these plugins?
  - Other Vaa3D plugins
    - Vaa3D-ITK plugins
    - Vaa3D-Bioformats plugins
- How to run plugins?
- How to write a plugin?
  - What you need before you start
  - How to compile a plugin from source code?
  - How to deploy a plugin binary?
- Advanced topic: what are exactly provided in the Vaa3D plugin interface?

What are Vaa3D plugins?

Vaa3D plugins are programs you can develop to take advantage of the management, visualization and analy
in C++ (most of the time), with Qt supported.

Current Status and Dev Roadmap

- Collaboration
- Remote [big] data access
- Better exploration of data
  - VR
  - AR
  - 3-D histogram of data distribution
  - More leveraging of virtual finger functions
  - Better support of ND-data on 2D-display devices
  - Exploration of ND-display devices
- Desktop -> laptop -> mobile platforms

- Collection of feedback from potential users