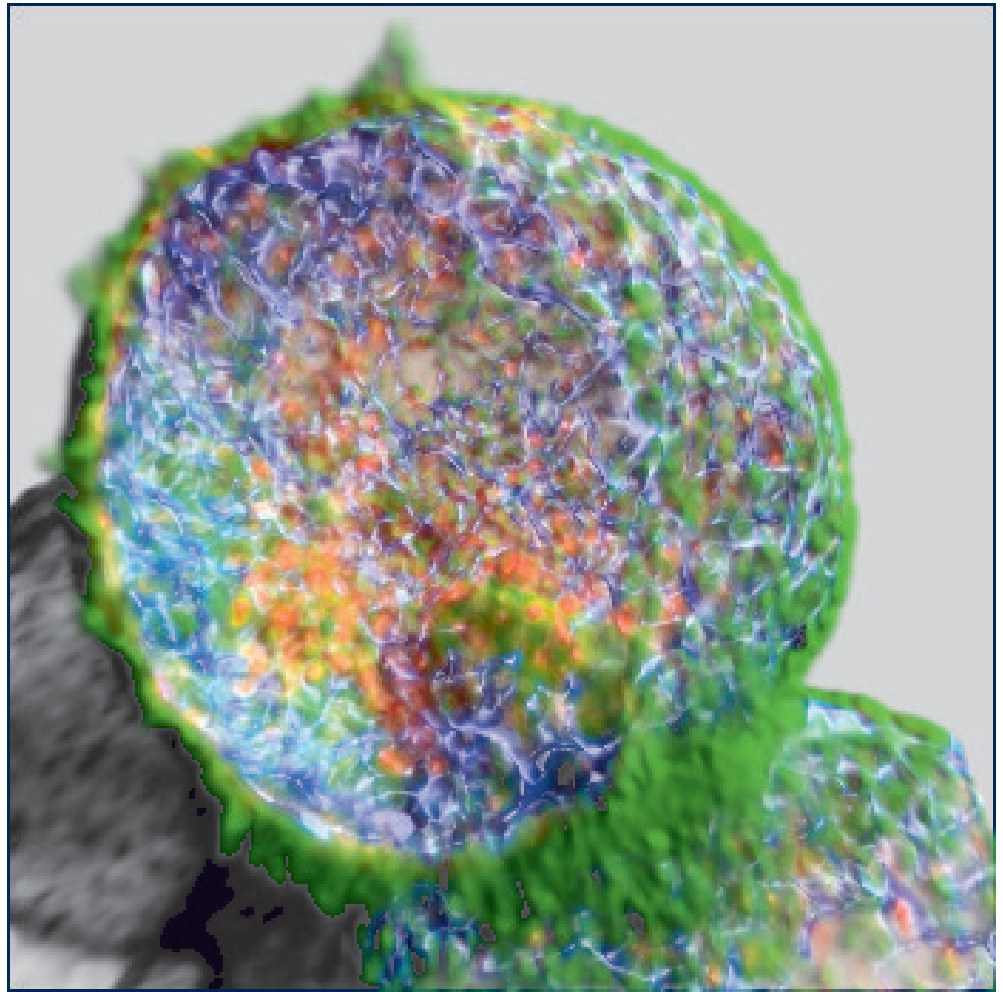


Huygens Essential



High quality deconvolution
visualization and analysis

Confocal ✧ Widefield ✧ Multi photon ✧ Nipkow disk
PSF distiller ✧ Volume and surface renderers
From 2D to multi channel 3D-time images

Windows ✧ Mac OS X ✧ Linux ✧ SGI Irix

Scientific Volume Imaging b.v.

www.svi.nl

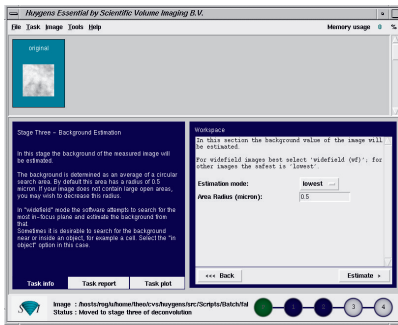


Startling clarity with Huygens Essential deconvolution

Huygens Essential makes high quality deconvolution available for everyone by combining state of the art deconvolution algorithms with a remarkable ease of use. Huygens Essential improves resolution and contrast in your microscopic images dramatically while effectively removing haze and noise. In this way structures and details become visible which would otherwise remain hidden.

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The **wizard-style** user interface guides you step by step through the deconvolution process.

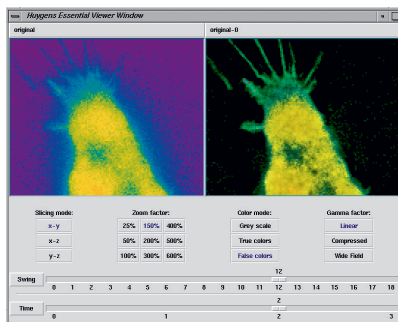


In the preprocessing stage the intelligent parameter checker scrutinizes the microscopic parameters. It marks suspicious optical conditions and warns you of under-sampling conditions. In the next stages you can use the automatic cropping tool,

inspect the image histogram to spot clipping or saturation and inspect the image background.

At the last stage you launch the iterative deconvolution run. During this stage **spherical aberration** is also corrected, as well as **bleaching** in the case of widefield images or time series. You can then launch the Twin Slicer by clicking on a thumbnail image to compare the deconvolution result with the recorded image. Alternatively, you can open multiple **volume rendering** windows to compare results.

You're in charge: whenever you are not fully satisfied with the result you can stop iterations and rerun with, for instance, a different background setting. Each run results in a different thumbnail image to keep track of your work.



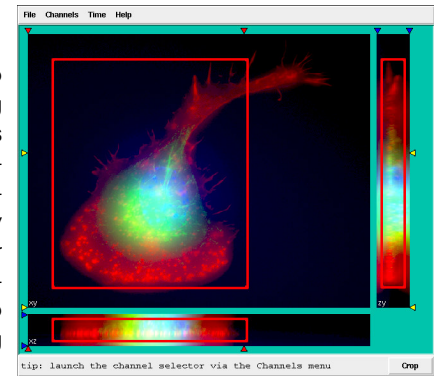
The **Twin Slicer** enables you to compare corresponding slices or time frames from the original and deconvolved image. You can simultaneously zoom and pan the images while selecting various contrast and colour

modes. You can swing through space or time and compare individual voxel values.

Multi channel images are handled by deconvolving the channels sequentially. When all channels are done you can select the best result on each channel to compose the final result.

Time series Huygens Essential is able to deconvolve time series of 3D or 2D images, automatically correcting for bleaching, drift and varying backgrounds.

The **Intelligent 4D cropping tool** allows you to trim images along four dimensions and to delete uninteresting channels. In that way you deploy your computer's processing power to only the interesting parts of the image.



Still, situations occur where your system's memory is not sufficiently large to allow deconvolution of an image as a whole. In these cases the image is split into **bricks**. The bricks are deconvolved one by one; the results seamlessly stitched together again.

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Automatic Point Spread Function. The imaging in a fluorescent microscope is completely described by its point spread function (PSF), which is therefore basic for the image restoration. The Huygens Compute Engine can accurately calculate a theoretical PSF based on a model of the microscope and known microscopic parameters. This ideal PSF will be automatically generated without any user intervention when no experimental one is provided, Huygens Essential does the calculation in the background without any notice.

Spherical aberration corrector. Any mismatch between the refractive indices of the objective and the preparation media will provoke spherical aberrations. When this happens, the theoretically calculated PSF automatically adapts to the sample depth to correct the aberrations as much as possible.

The PSF distiller is the interactive tool present in Huygens Essential that allows you to obtain an experimental point spread function from imaged calibration beads. A measured PSF will take into account deviations in your optical device from the ideal conditions, improving the quality of the restorations.

Batch Processor. Once you know how to deal with a particular kind of dataset you can restore a couple or more of similar datasets automatically. This is called batch processing. Huygens Essential includes an interactive batch tool that will allow you to easily restore large collections of images, even with different parameters or PSF's.

A complete list of features is available at www.svi.nl/products.



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A free demo copy of Huygens Essential can be downloaded from our website www.svi.nl.

Volume and surface visualization with Huygens Essential

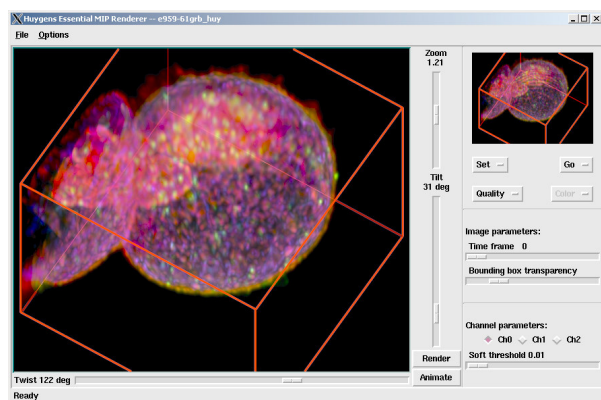
Huygens Essential interactive tools for volume and surface rendering easily let you obtain striking high-resolution images for press or on-line publishing. Eye-catching animations for presentations or web pages

can also be easily rendered by automatically changing the viewpoints or visualization parameters in a smooth way. The renderers work very fast and without needing any special graphic card.

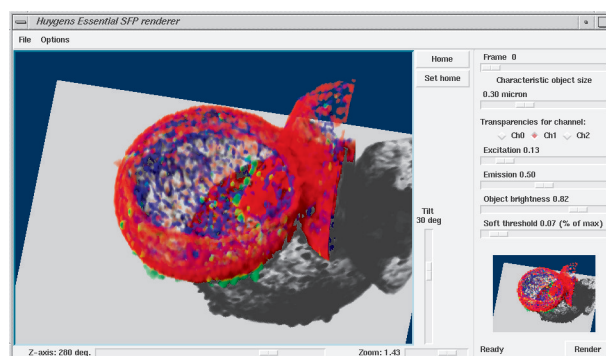
Scientific Volume Imaging has a long experience in visualization next to its strong focus on deconvolution. We developed visualization tools that were part of Huygens Professional, FluVR or FreeSFP. Since the year 2000 this has resulted in adding a set of powerful visualization tools to the Huygens Essential basic as well, that keeps growing nowadays.

Maximum Intensity Projection (MIP) enables you to obtain a spatial projection of your 3D microscopy data from the pointview you wish.

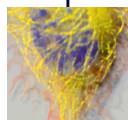
The renderer projects, in the visualization plane, the voxels with maximum intensity that fall in the way of parallel rays traced from the viewpoint to the plane of projection. This is a very fast visualization technique that is usually combined with viewpoint rotation to make animations.



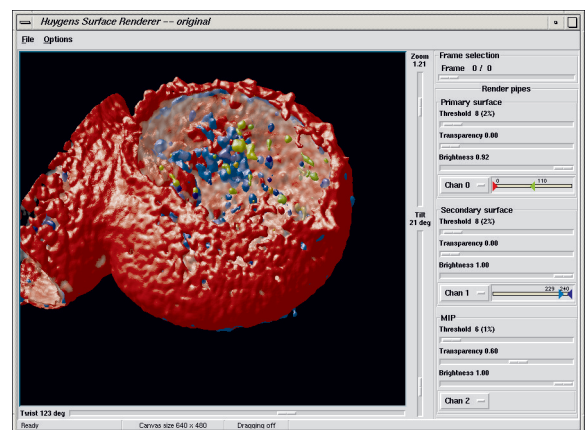
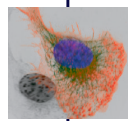
Volume Rendering. Based on the **Simulated Fluorescence Process** algorithm (SFP), the volume renderer takes the 3D microscopy image as a distribution of fluorescent material, simulating what happens if the material is excited and how the subsequently emitted light travels to the observer.



- Depth cue rich physically realistic algorithm
- Interactive manipulation of viewpoint, transparencies and zooming
- 4D support & animation
- Ray-tracing based, it does not require any special graphical board



Surface Rendering. Introduced as an optional extra visualization tool, it allows you to explore easily the different objects present in your data.



- Fast raytracing rendering.
- No need for any special graphic card as would be necessary for conventional polygon based techniques.
- Three graphic pipes available to visualize your image's data channels: two surface pipes and one MIP pipe, enabling you to mix Surface and MIP together.
- Independent control of transparency and brightness in each graphic pipe.



The Twin Slicer: Instant comparison of your restored and original dataset, or of different slices in your image (see previous page).

More...

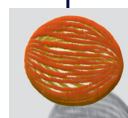


Image I/O file formats. Huygens Essential is able to deal with images in a variety of formats:

- Reads/writes ICS (Image Cytometry Standard), Nikon-ICS, Leica style TIFF series, Biorad 'pic', and Imaris classic images
- Reads Zeiss 'Lsm5', Metamorph 'stk', 'MRC', Olympus 'Fluoview', DeltaVision 'IMSsubs'
- Reads/writes a single or numbered series TIFF images into/from 3D volume image
- **4D and multichannel support:** ICS, TIFF series, Biorad, numbered 'stk'



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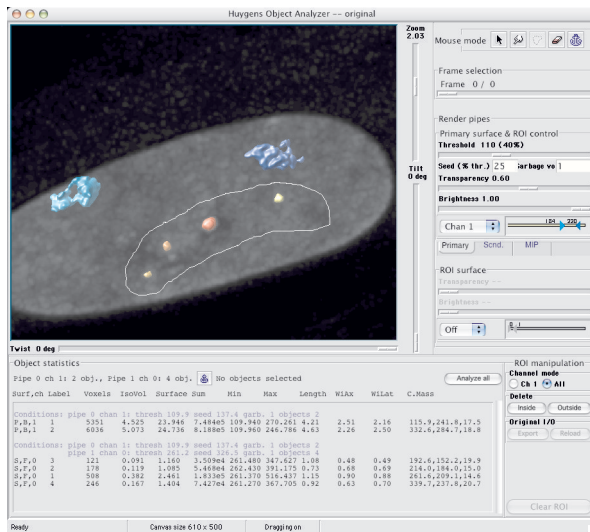
On-line knowledge and support resources
with much more information at support.svi.nl

Huygens Essential interactive volume data analysis

Huygens Essential optional tools for interactive colocalization and object analysis combine effective 3D data manipulation, selection and visualization with powerful characterization algorithms. These can be

configured or redefined by the user at will, because Huygens Software uses the widely spread Tcl environment as scripting language, expanded for easily handling multidimensional images.

The Object Analyzer optional tool in Huygens Essential allows you to interactively obtain statistics of individual objects by clicking on them, or analyzing all objects with a single button press.

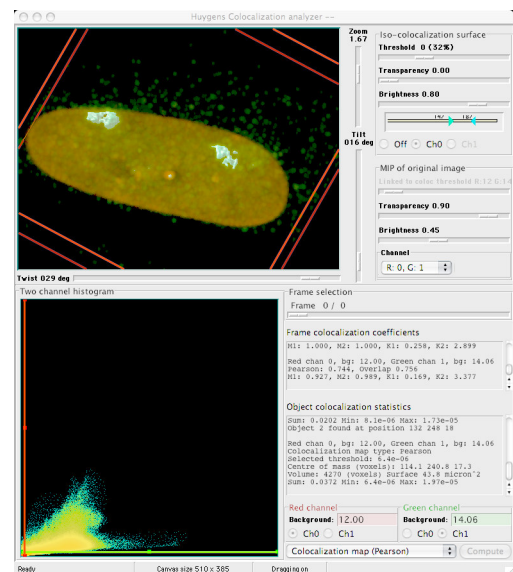


The image is segmented into objects by an effective seed-threshold level and connection technique. To remove too small objects in an early stage from the analysis a garbage level can be set below which objects are discarded. After that, detected objects are automatically labeled and sent to a continuous surface renderer.

The Colocalization Analyzer allows you to obtain quantitative information about the amount of spatial overlap between structures in different data channels, for 3D images and 3D-time series. As this overlap can be defined in many ways, the colocalization analyzer gives you the colocalization coefficients most commonly used in literature: Pearson, Overlap, and Manders M and K.

One of the features of the colocalization analyzer is **iso-colocalization object analysis**, that allows you to quickly determine the properties of the different colocalization regions in your data. This is realized by visualizing the colocalization map as iso-colocalization surfaces. In the colocalization analyzer these surfaces are computed at the same time as the coefficients

The surface objects show regions in which the degree of colocalization exceeds a certain value. By clicking on the objects local colocalization parameters are computed and reported. To relate the iso-colocalization objects to the original data the surface objects can be blended with a MIP projection of the data. The color range in which these objects will be displayed can be modified using a Hue Selector



More...

Multiple platforms. Huygens Essential runs on PCs, Macs and workstations with:

- Windows NT, 2000, Server 2003, XP
- Apple Mac OS X
- Linux

When your datasets are so large that they outgrow 32 bit platforms you can always run **64 bit multiprocessor** versions of the Huygens Software.

Contact and Support. Learn more on microscopy and how our software can help you with your research on the SVI-wiki: support.svi.nl/wiki.

When you would like to stay informed on our latest developments, simply send an email to sales@svi.nl with the subject: 'Announcements'.

Cover illustration and 'Huygens Visualization Tools' images: isolated Rat Hepatocyte couplet recorded by Dr. Permin Marbet at the Department of Anatomy, University of Basel, Switzerland (head: Prof. Lukas Landmann) as deconvolved with Huygens and visualized with the spectral SFP volume rendering algorithm, the surface renderer, and the maximum intensity projector. Cropper illustration and first three color images between text columns: macrophage fluorescently stained for tubulin (yellow/green), actin (red) and the nucleus (DAPI, blue). Recorded by Dr. James Evans, Whitehead Institute, MIT, Boston MA, USA, using widefield microscopy. Twin Slicer illustration shows 3D-time series of a similar macrophage, also recorded by Dr. James Evans. Fourth color image in columns separator is a 25 µm pollen grain *Spathiphyllum* recorded by Dr. Erik Manders, SILS, University of Amsterdam. 'Huygens Analysis Tools' images: cell nucleus FISH-stained recorded by Dr. Sandra Goetze at the Nuclear Organization Group, SILS, University of Amsterdam (head: Prof. Roel van Driel). Greyscale pairs between text columns: original and deconvolved widefield image: Rhodamine phalloidin labelled SW480 cell derived from a colorectal adenocarcinoma. Prepared by Dr. P. Roux and G. Gaddea, recorded by Dr. Pierre Travo, Centre de Recherches de Biochimie Macromoléculaire, Montpellier, France.

