



Alone in the dark making big data

luke hammond / QBI

Mouse Hippocampus

Nika Mohannak / Meunier Lab

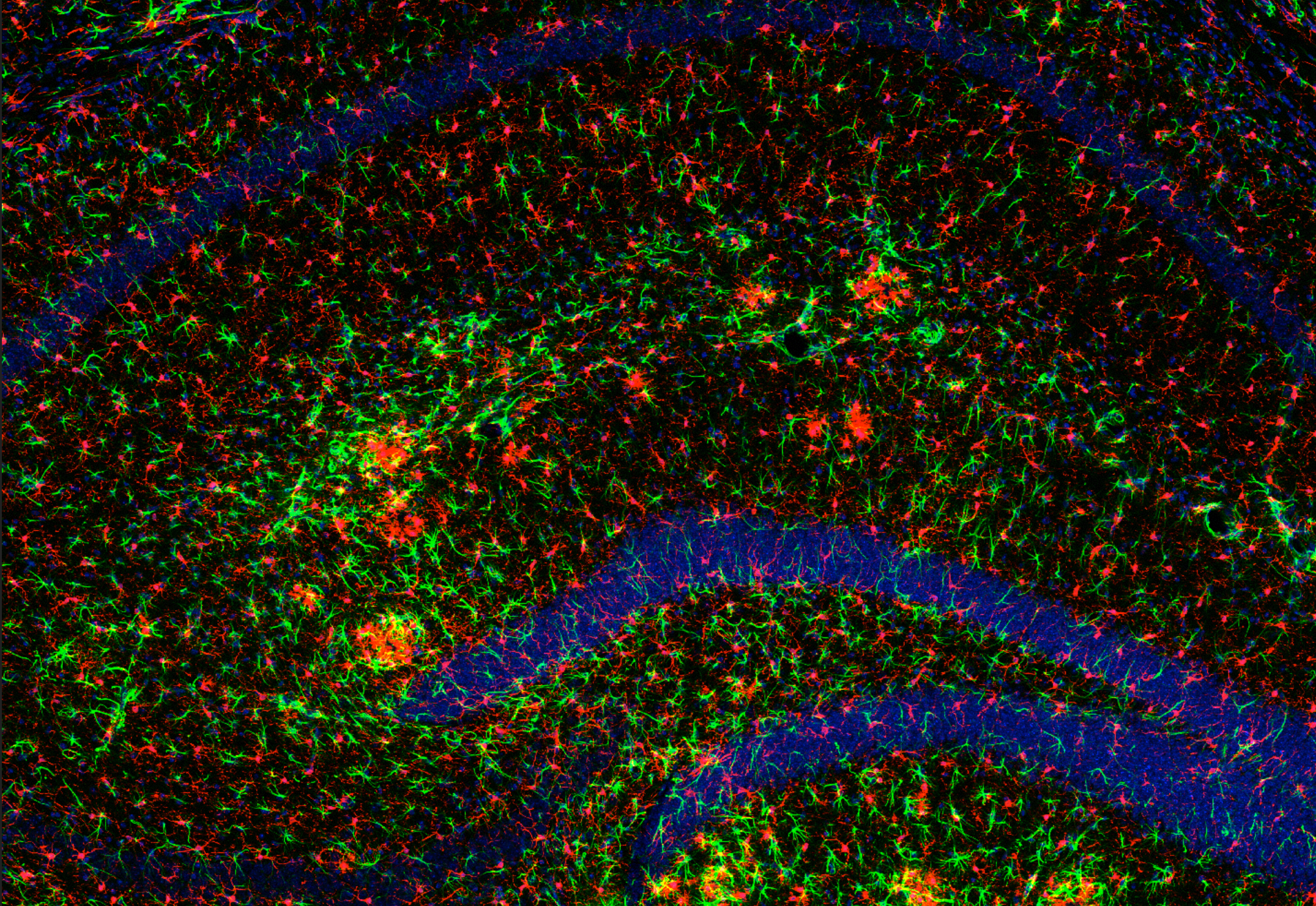
Axiolmager + ApoTome

20x 0.8 NA Water Objective

0.3 x 0.3 μm XY

1.2 Z step size

12 slices x 6 tiles



Zebrafish / GCaMP6

Dr. Jeremy Ullmann

Yokogawa W1 SDC

40x 1.2 NA Water Objective

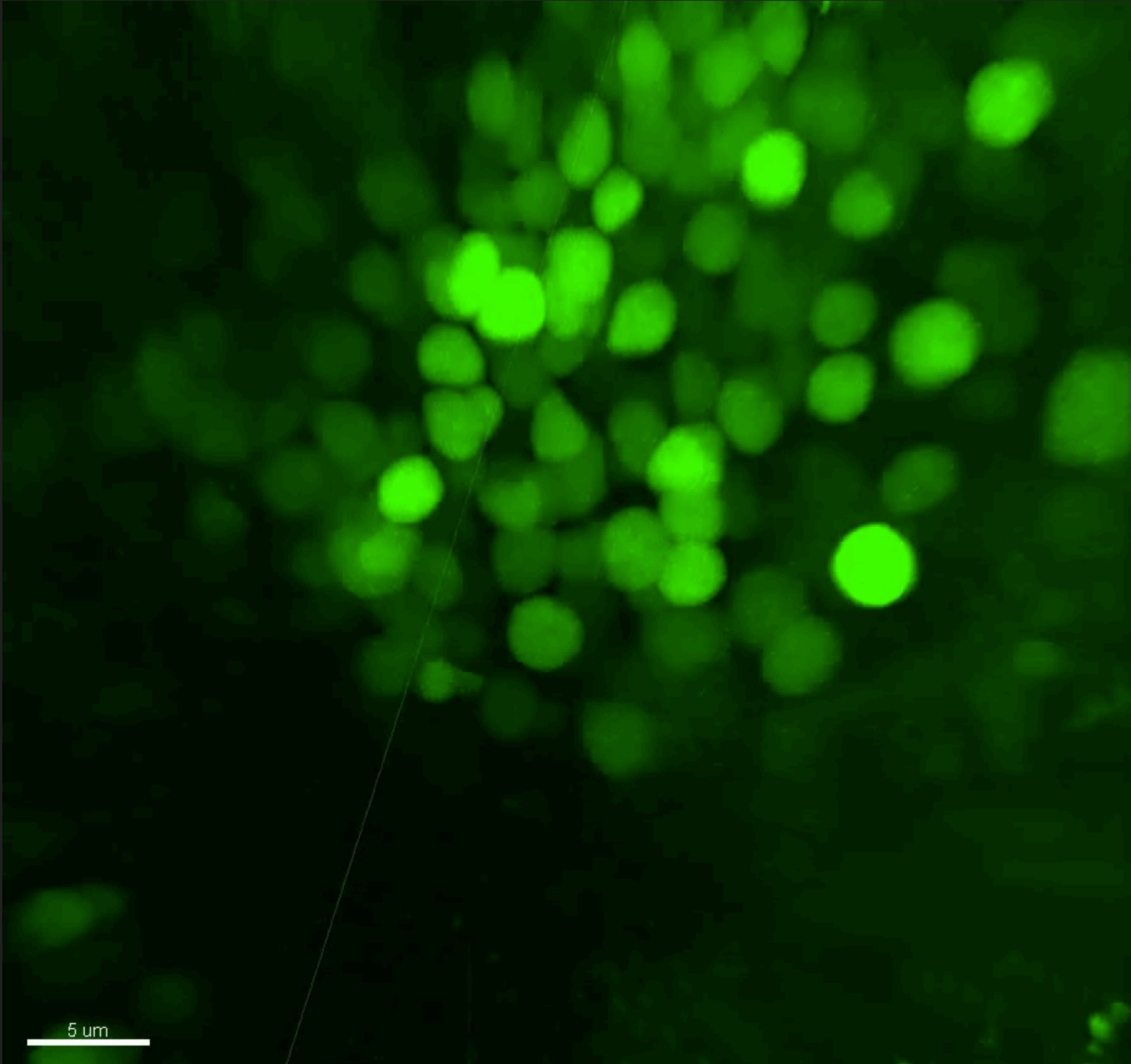
0.15 x 0.15 μm XY

0.33 Z step size

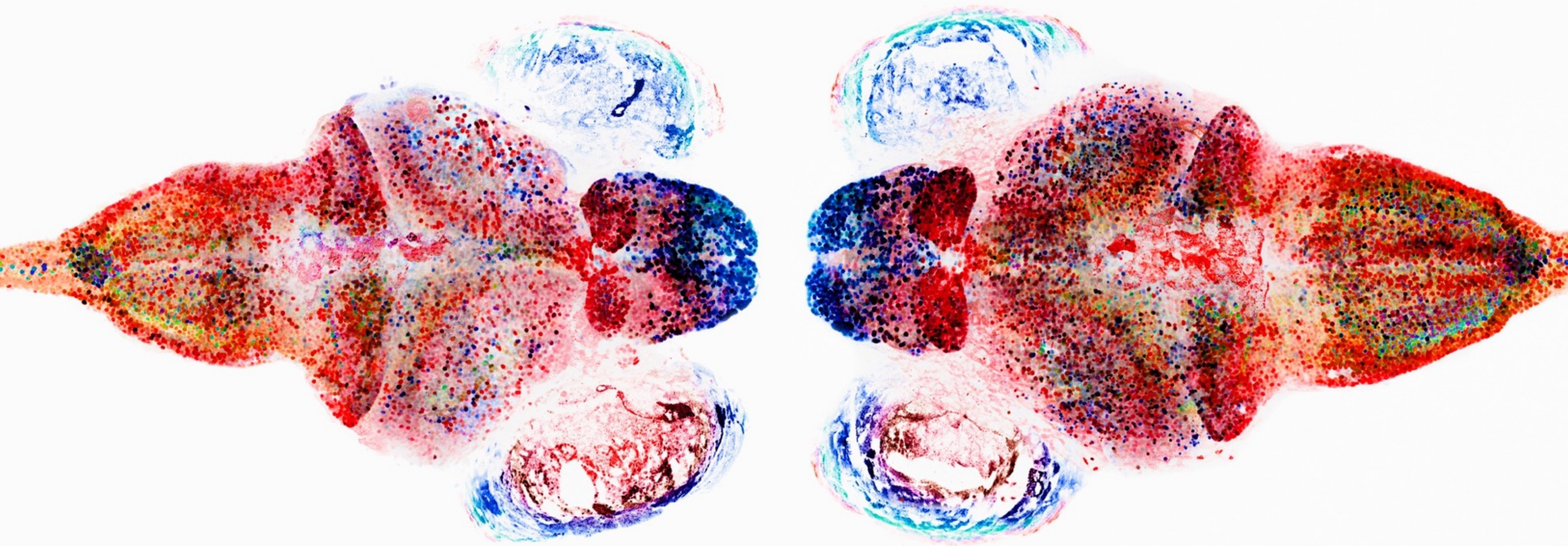
800 slices x 6 tiles

20 minutes acquisition

40 GB data set



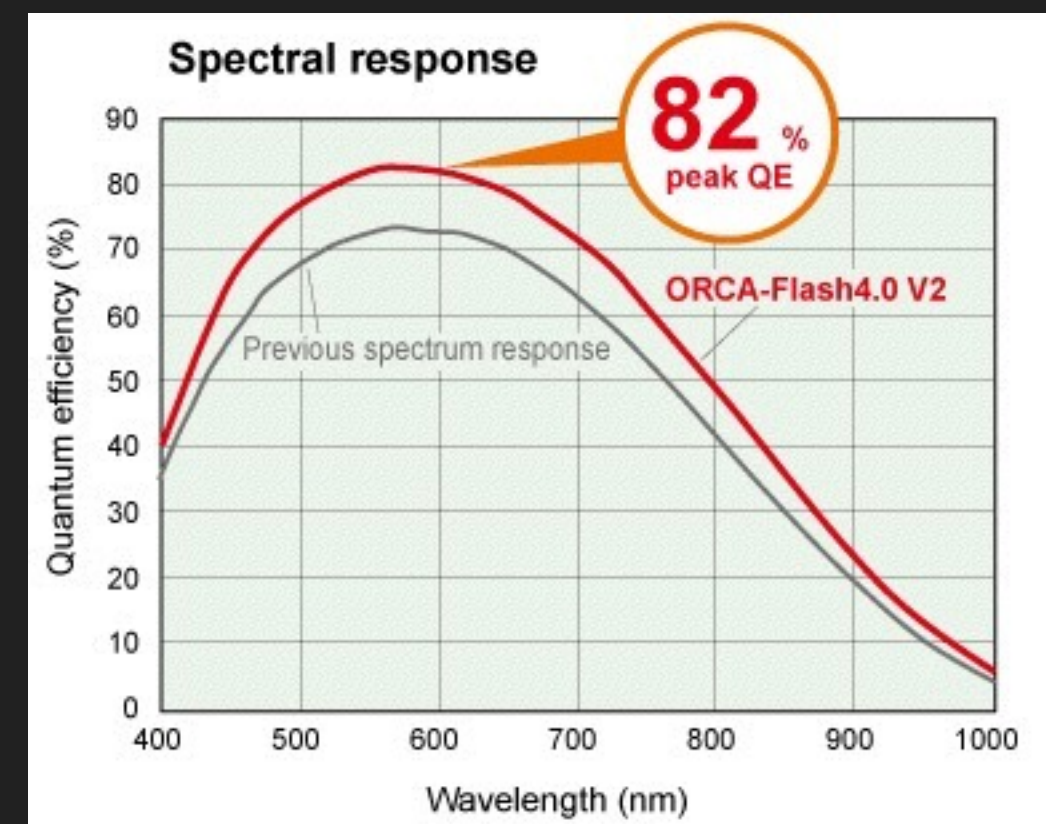






sCMOS Cameras

Hamamatsu Flash 4v2 high QE / 2015



100 frames / second

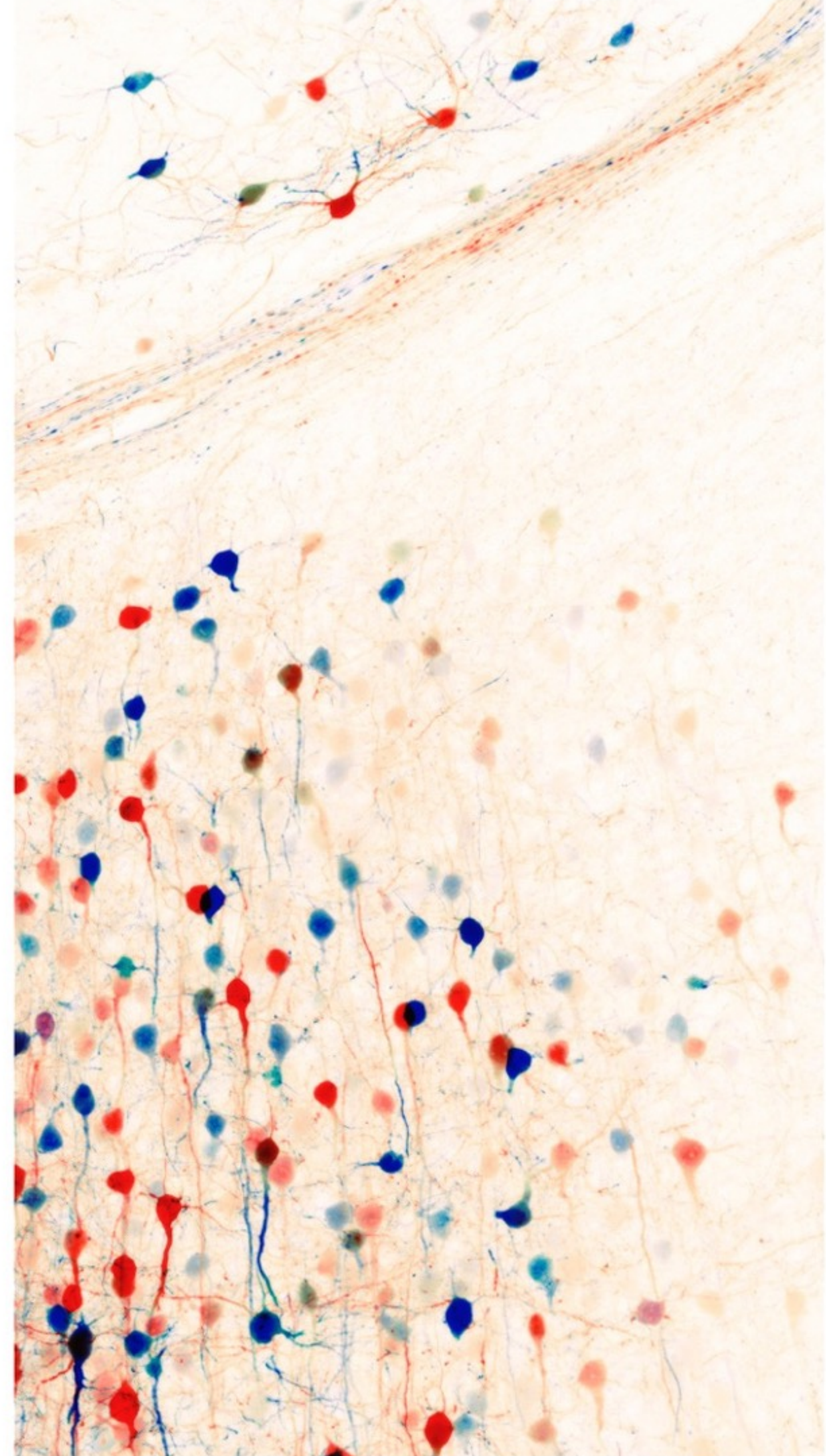
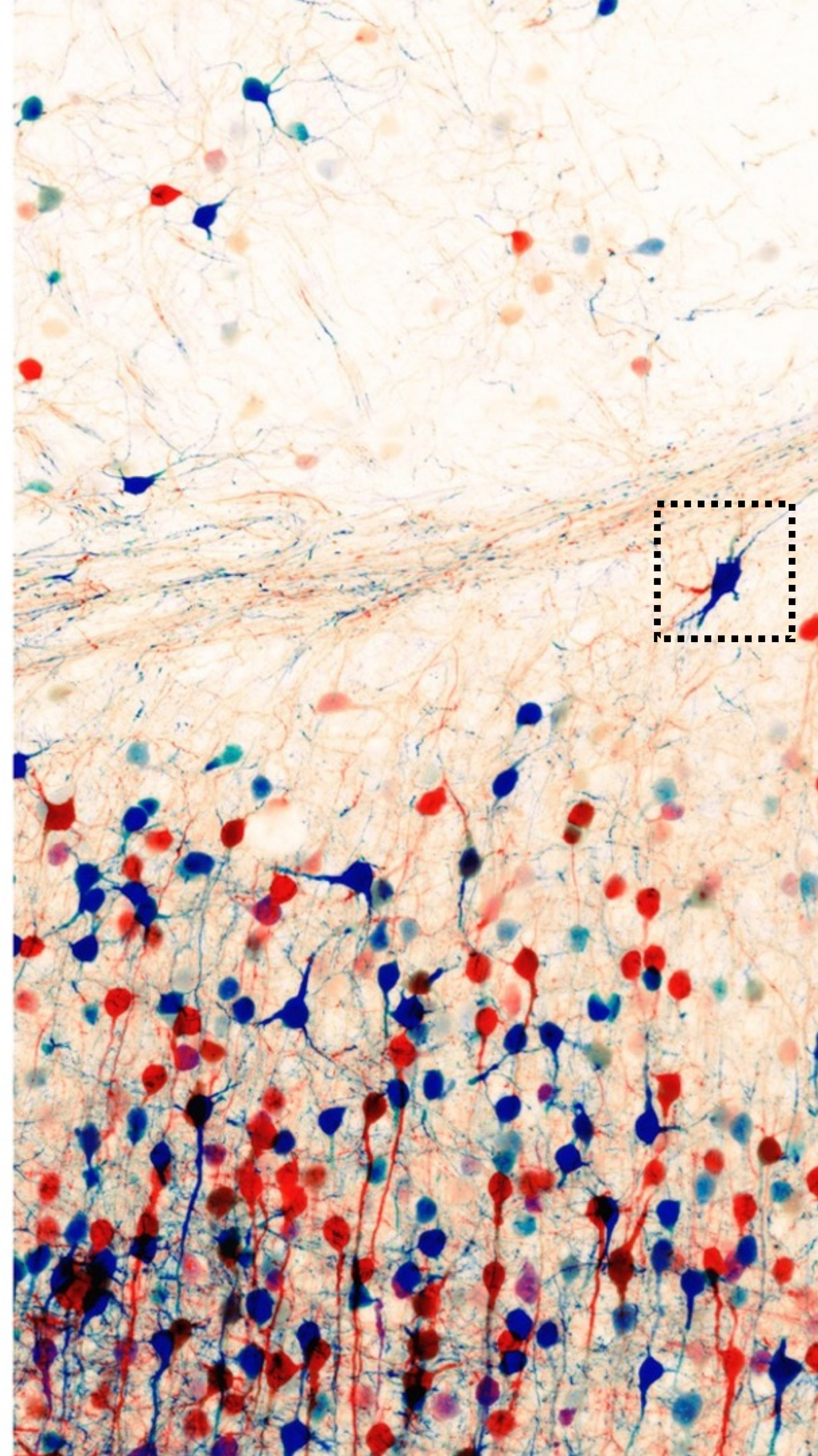
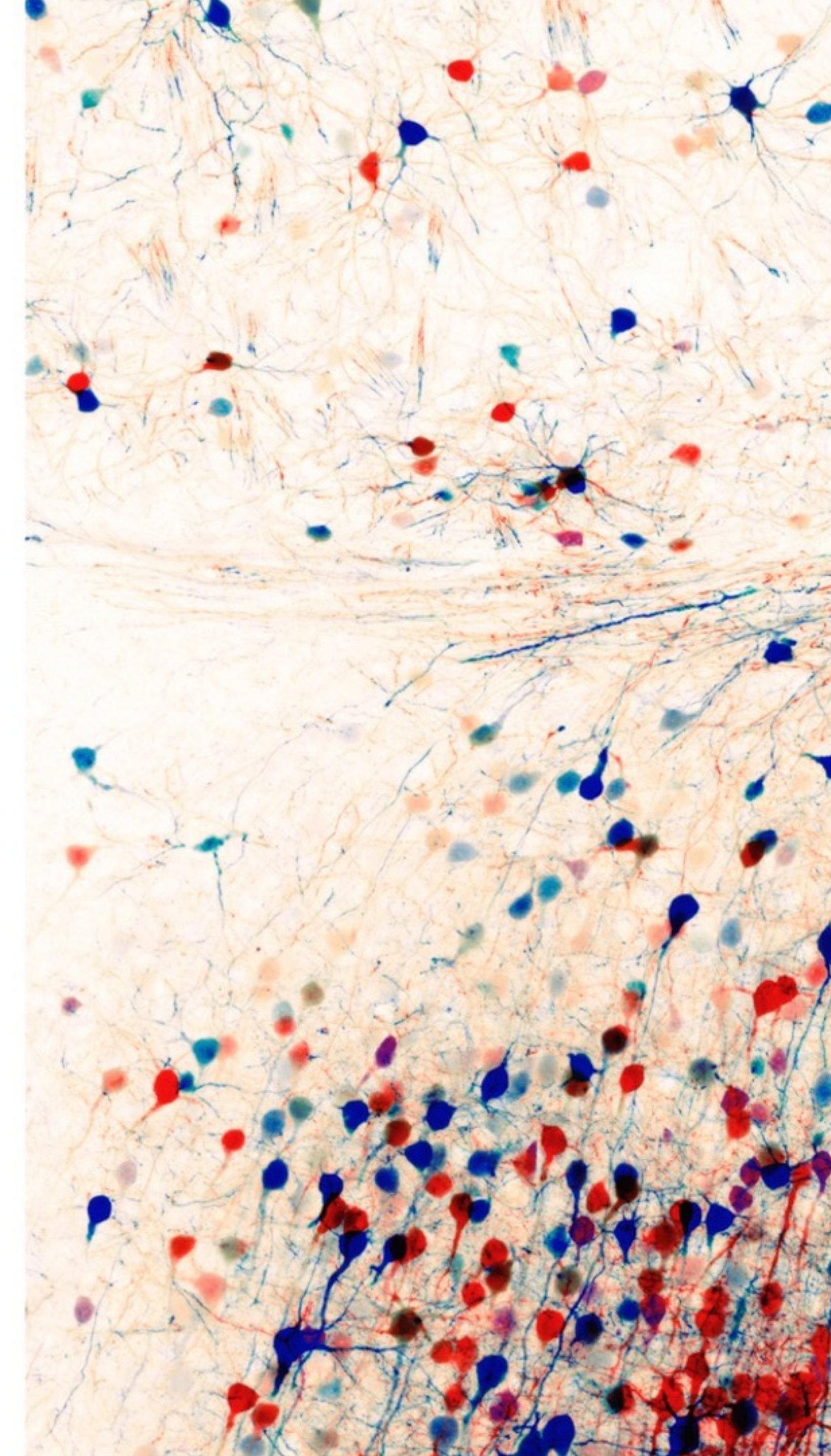
10 MB / frame

=

1GB / second



PMT = ~20%
GaAsP = ~45%



LSM 710

63x 1.4 NA Oil Objective

0.64 x 0.64 μm XY

0.13 μm Z step size

180 slices x ~20 μm

1024 x 1024 pixels

scan speed 7 (1.58 μs)

1 frame = 4 seconds

z stack = 12 minutes

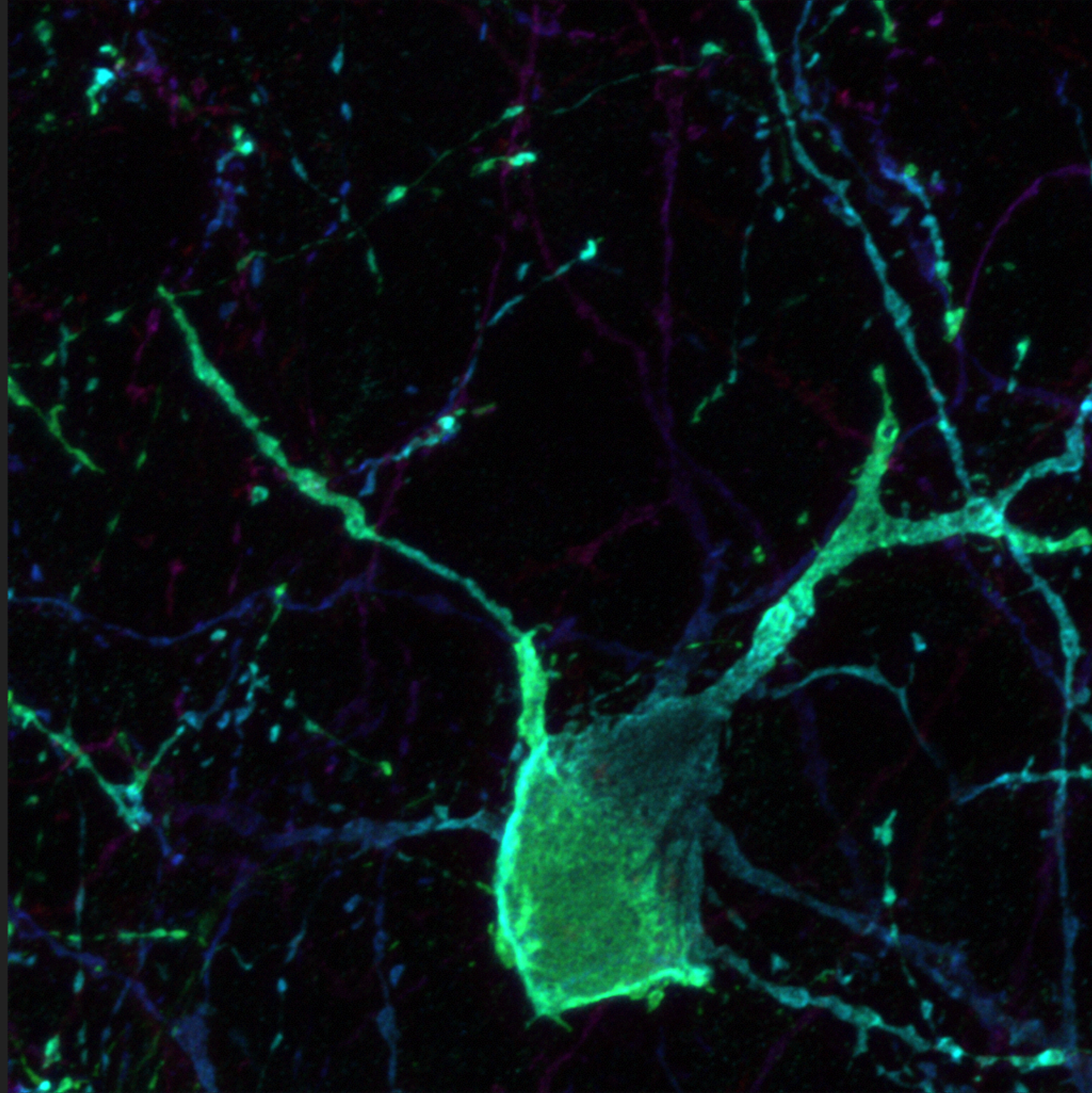
total size = 360MB

Or

0.4 z step

z stack = 4 minutes

total size = 120MB



Yokogawa W1

100x 1.4 NA Oil Objective

0.64 x 0.64 μm XY

0.13 μm Z step size

180 slices x ~20 μm

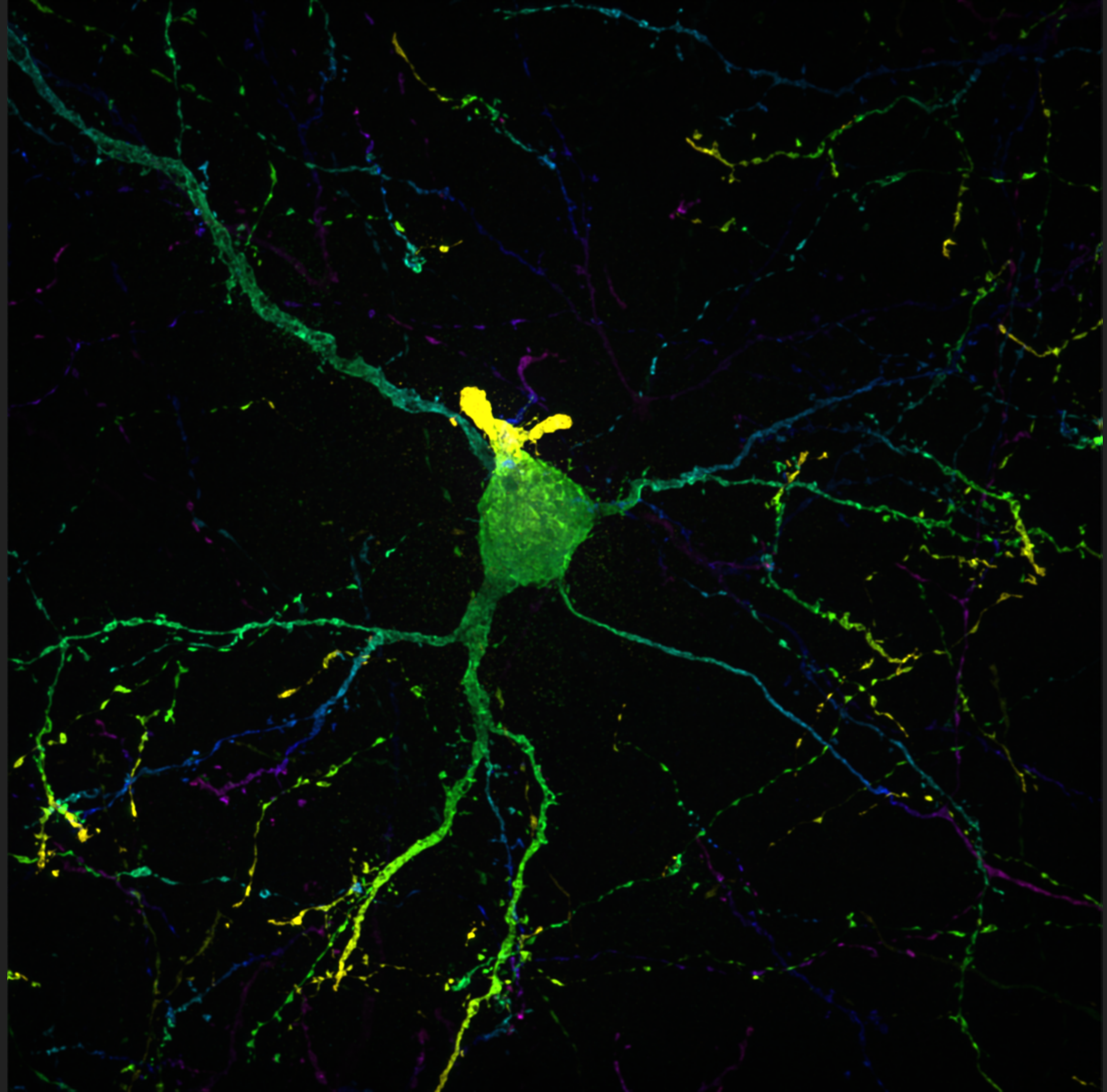
2048 x 2048 pixels

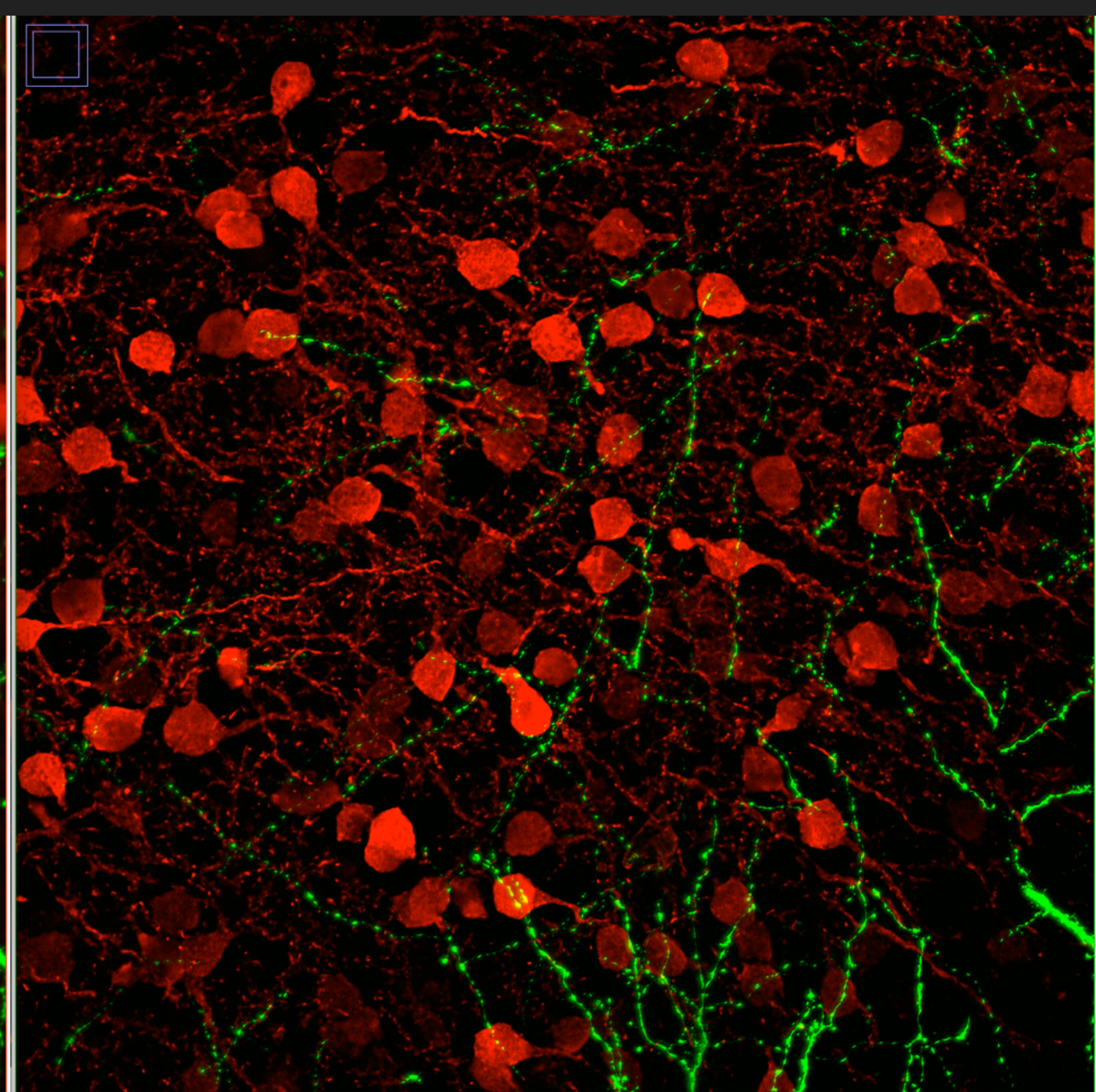
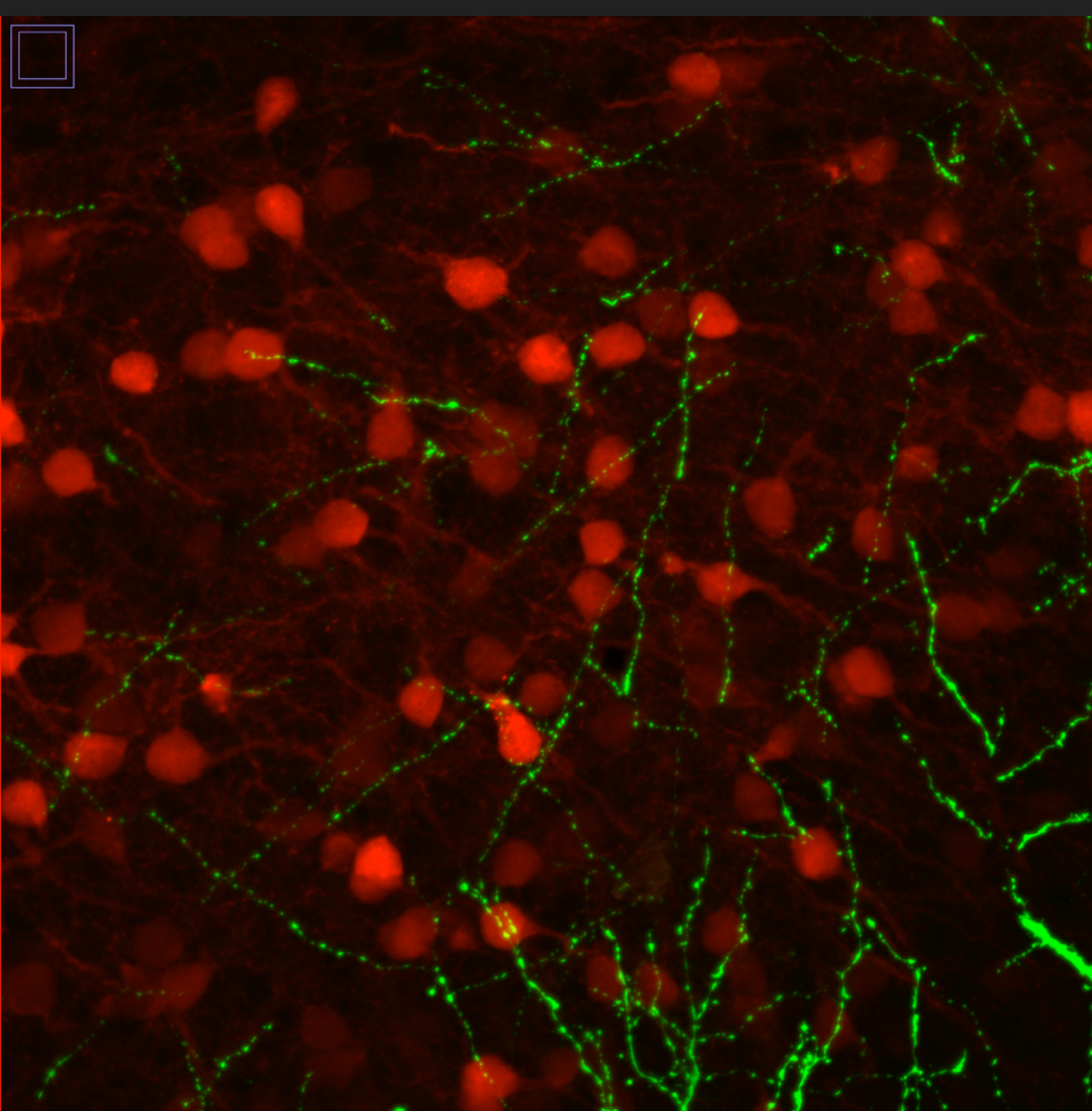
100msec exposure

1 frame = 0.1 seconds

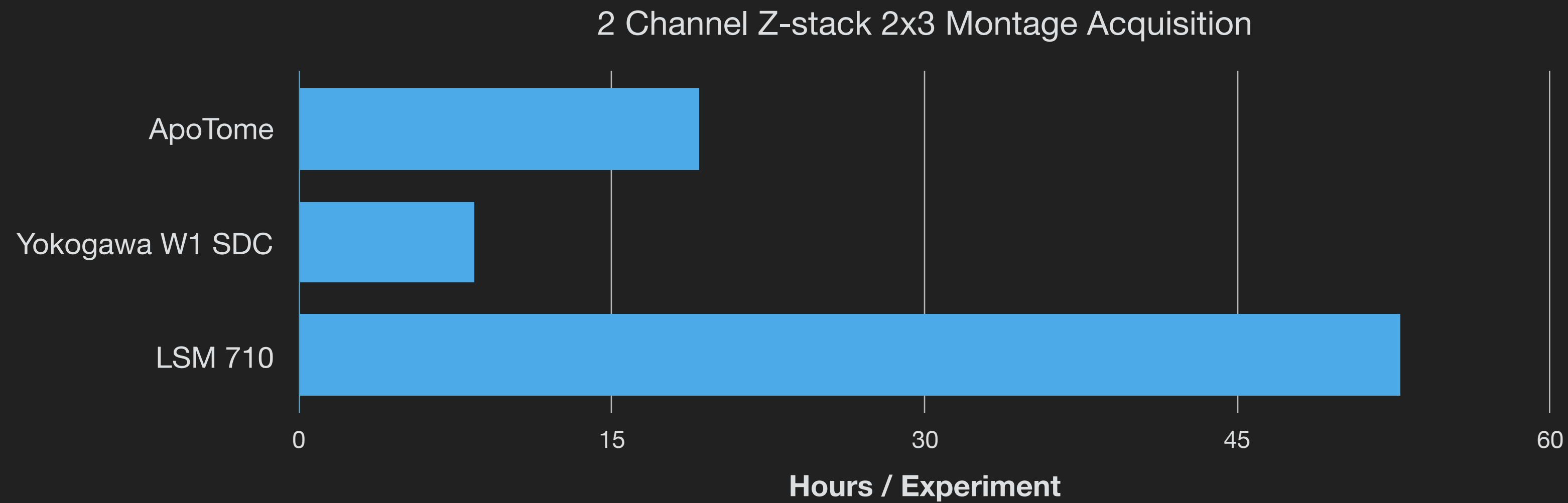
z stack = 50 seconds

total size = 1.5GB





Diskovery 20x
2048 x 2048 0.3 μ m

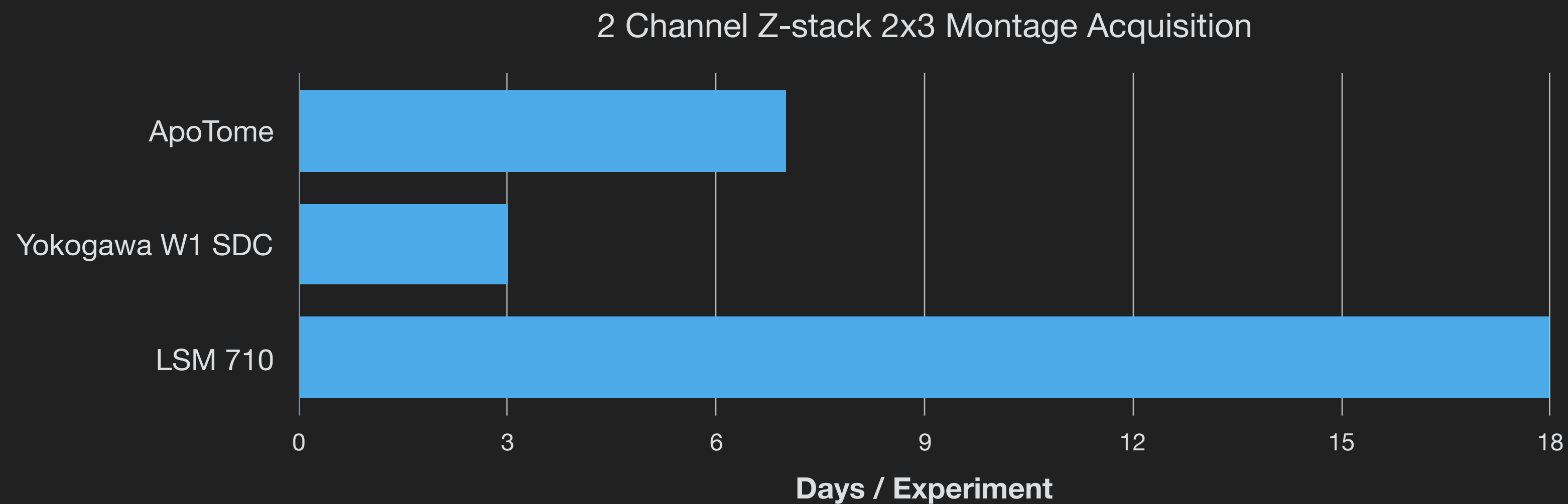


Assuming 2 Conditions

With 6 Animals / Condition

Imaging 12 tissue sections with a 2x3 montage and 15 slices

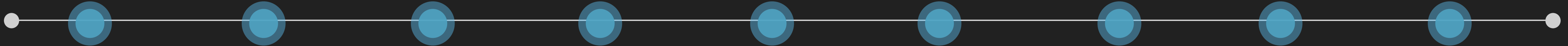
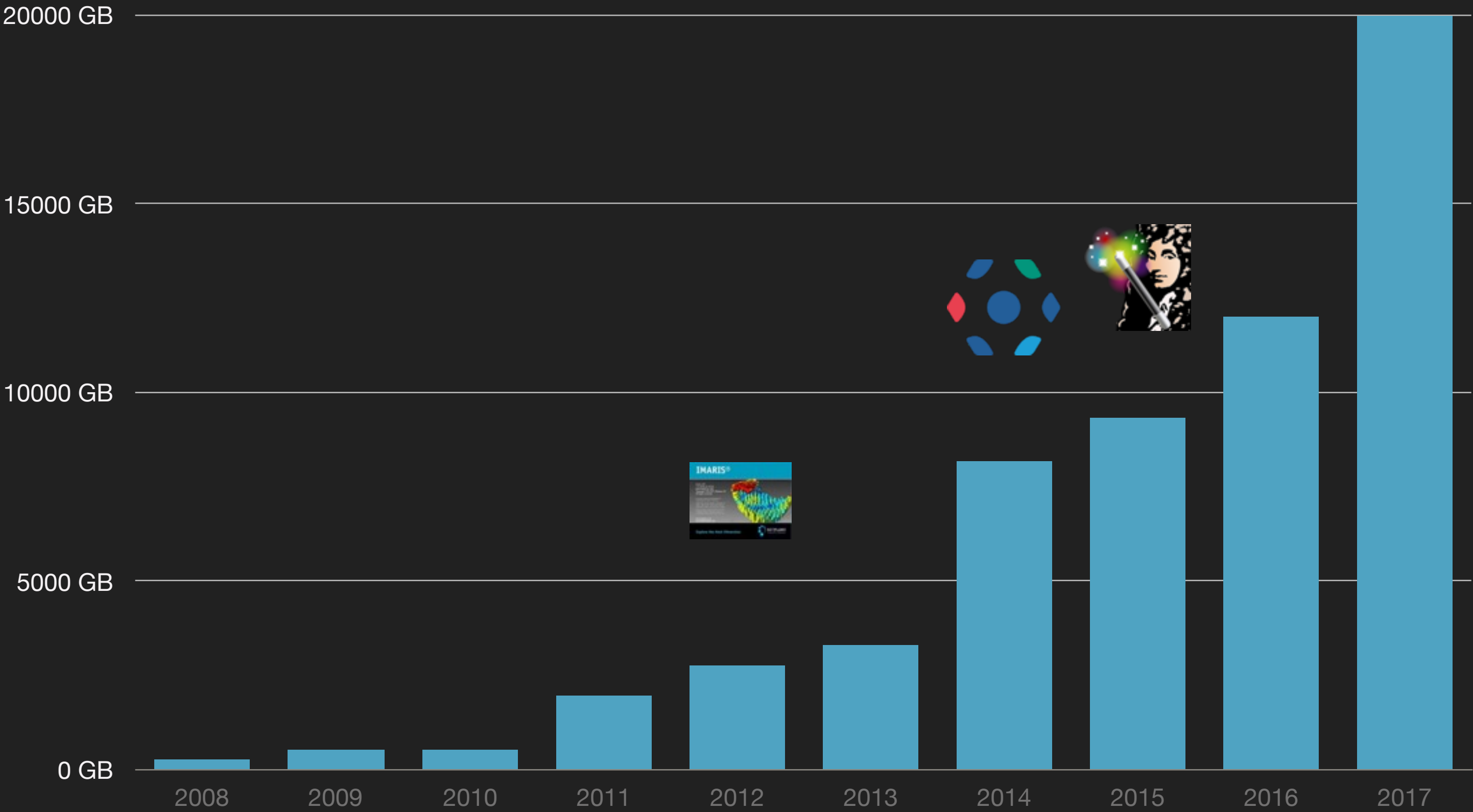
2 channels / 300 msec exposure



Assuming 3 hours of imaging each day

QBI Microscopy Facility Growth

Data generation
GB /month



2008

2009

2010

2011

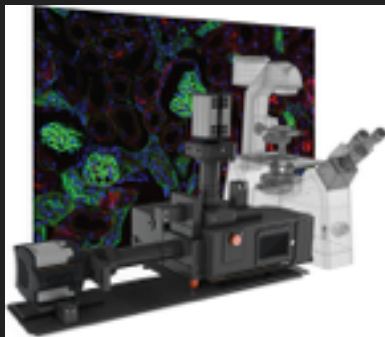
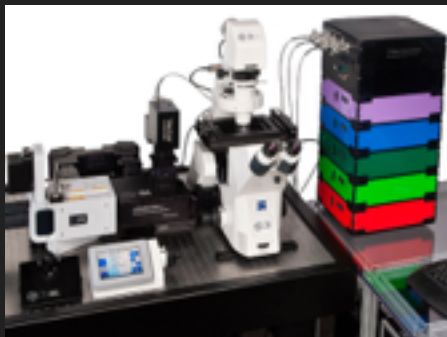
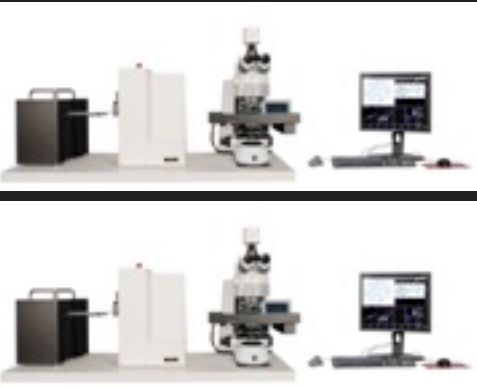
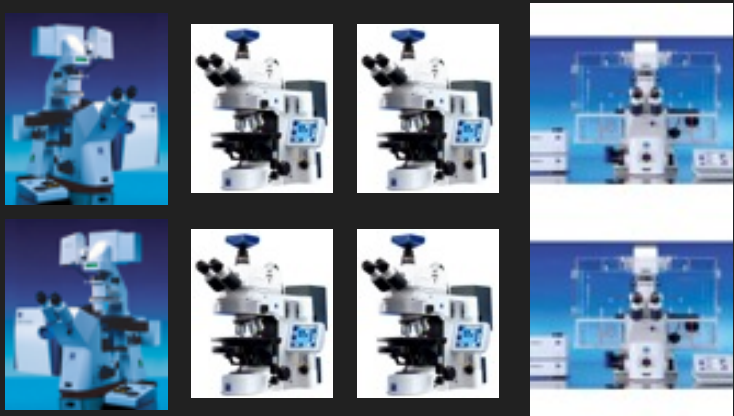
2012

2013

2014

2015

2016



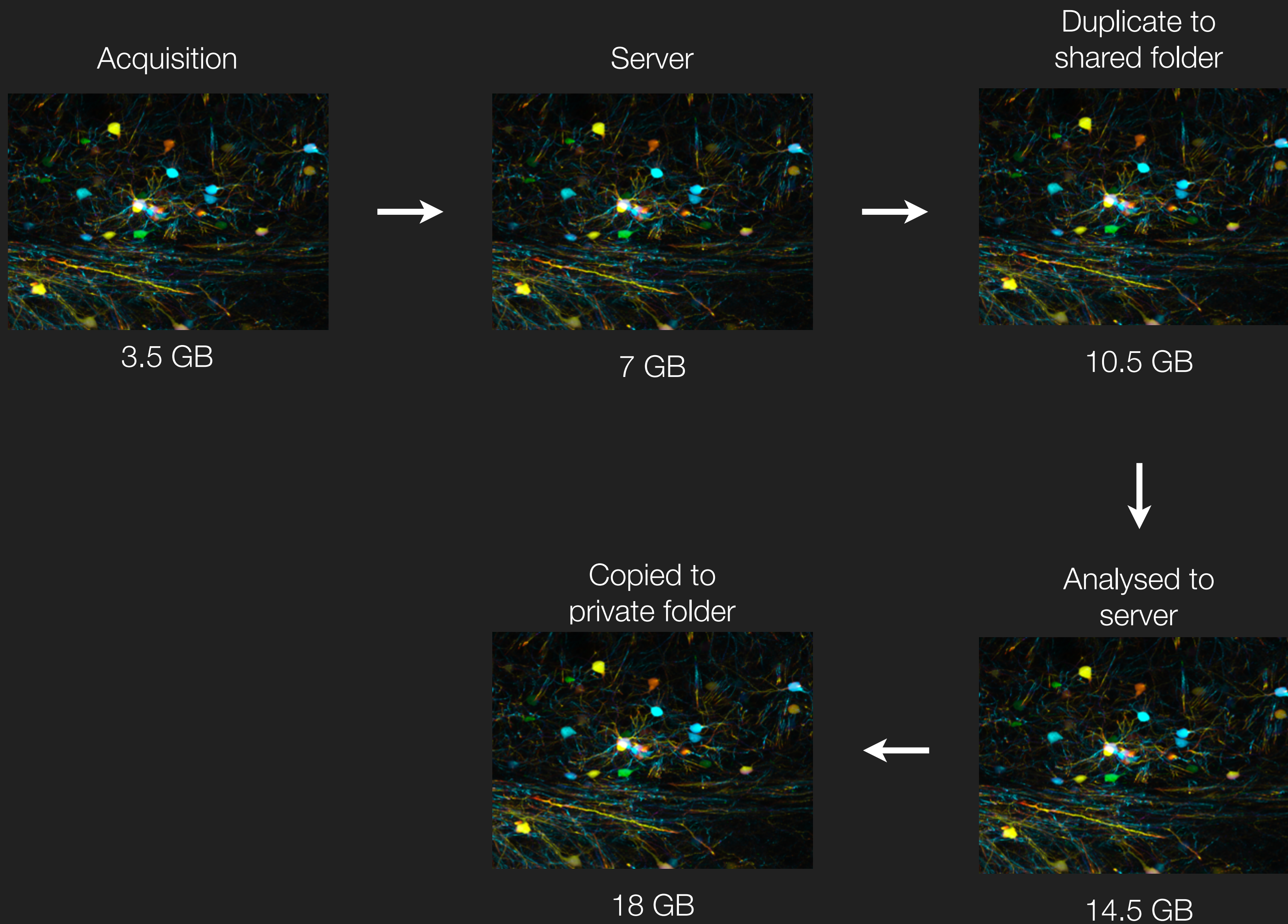
For assistance please contact

Network > home.qbi.uq.edu.au > group_microscopy > QBI Slide Scanner Images > FL Slide Scanner

Organize Burn New folder					
Search FL Slide Scanner					
Favorites Desktop Downloads Recent Places Libraries Documents Music Pictures Videos Computer Imaris Workstation (C:) Temporary Data (D:) Network					
Name	Date modified	Type	Size		
13a-8-401m3 001-2~B	15/03/2012 12:03 ...	Imaris File	8,369,572 KB		
13a-8-401m3 001-3~B	15/03/2012 4:09 AM	Imaris File	7,075,789 KB		
13a-8-401m3 001-4~B	15/03/2012 8:26 AM	Imaris File	7,366,319 KB		
13a-8-401m3 001-5~B	15/03/2012 12:49 ...	Imaris File	7,647,710 KB		
13a-8-401m3 002-1~B	15/03/2012 5:23 PM	Imaris File	7,861,739 KB		
13a-8-401m3 002-2~B	15/03/2012 10:05 ...	Imaris File	8,072,823 KB		
13a-8-401m3 002-3~B	16/03/2012 2:36 AM	Imaris File	7,673,486 KB		
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13a-8-401m3 002-5~B	16/03/2012 11:50 ...	Imaris File	7,220,529 KB		
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13a-8-401m3 003-5~B	27/02/2012 5:50 AM	Imaris File	6,043,247 KB		
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13a-8-402m1 001-3~B	25/02/2012 2:25 AM	Imaris File	6,827,168 KB		
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13a-8-402m1 002-3~B	26/02/2012 12:14 ...	Imaris File	7,171,454 KB		
13a-8-402m1 002-4~B	26/02/2012 4:37 AM	Imaris File	7,445,422 KB		
13a-8-402m1 002-5~B	26/02/2012 9:08 AM	Imaris File	7,668,908 KB		
13a-8-402m1 003-1~B	26/02/2012 1:41 PM	Imaris File	7,604,048 KB		
13a-8-402m1 003-2~B	26/02/2012 6:12 PM	Imaris File	7,527,479 KB		



1,382 items status: Online
Offline availability: Not available



1. Need to be able to preview image data before accessing it
2. Data preservation when researchers leave lab
3. Prevent unnecessary duplication of image data
4. Facilitate server based analysis pipelines and deter ad hoc manual approaches
5. Reanalysis and sharing of analysis tools





OMERO @ QBI

Polaris

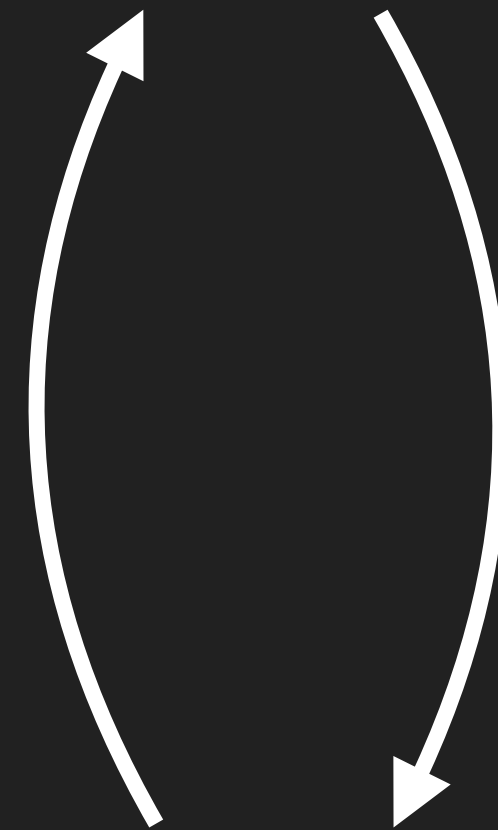
 **nectarcloud**

 **openstack**

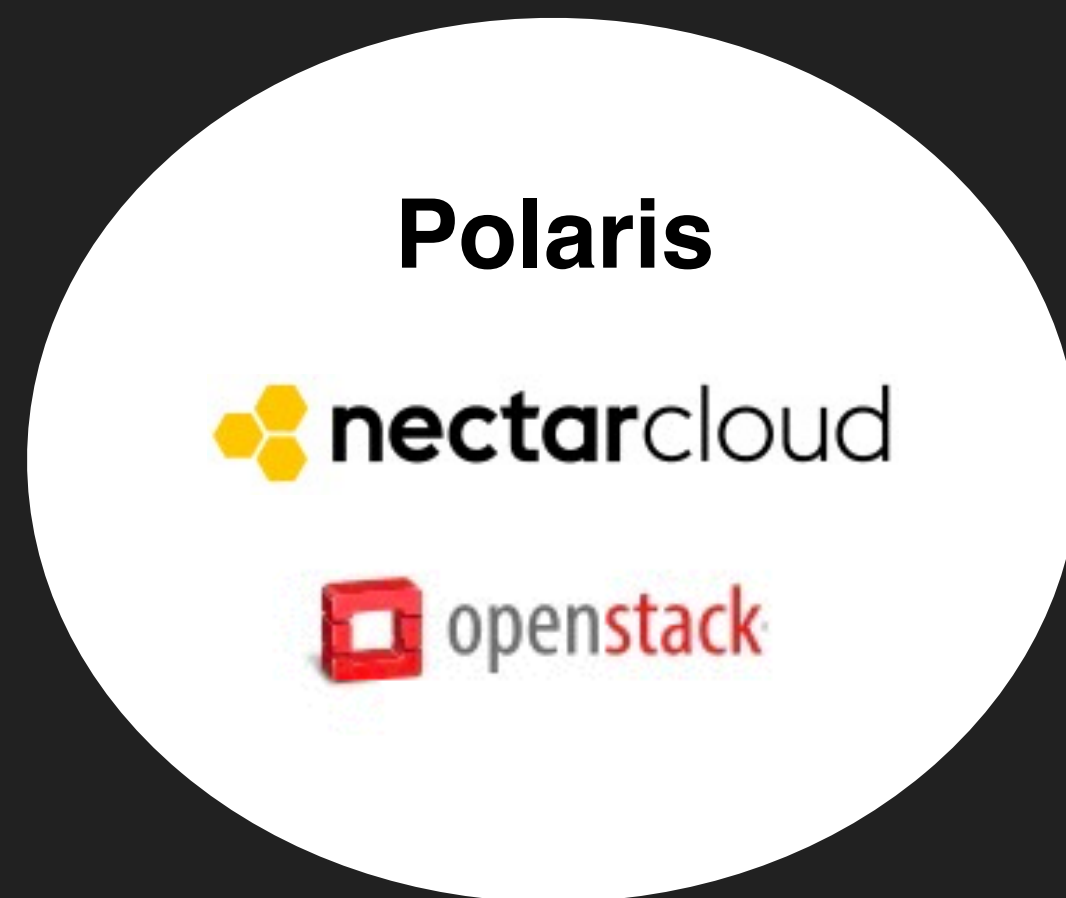


16 VCPU
64GB RAM

OMERO

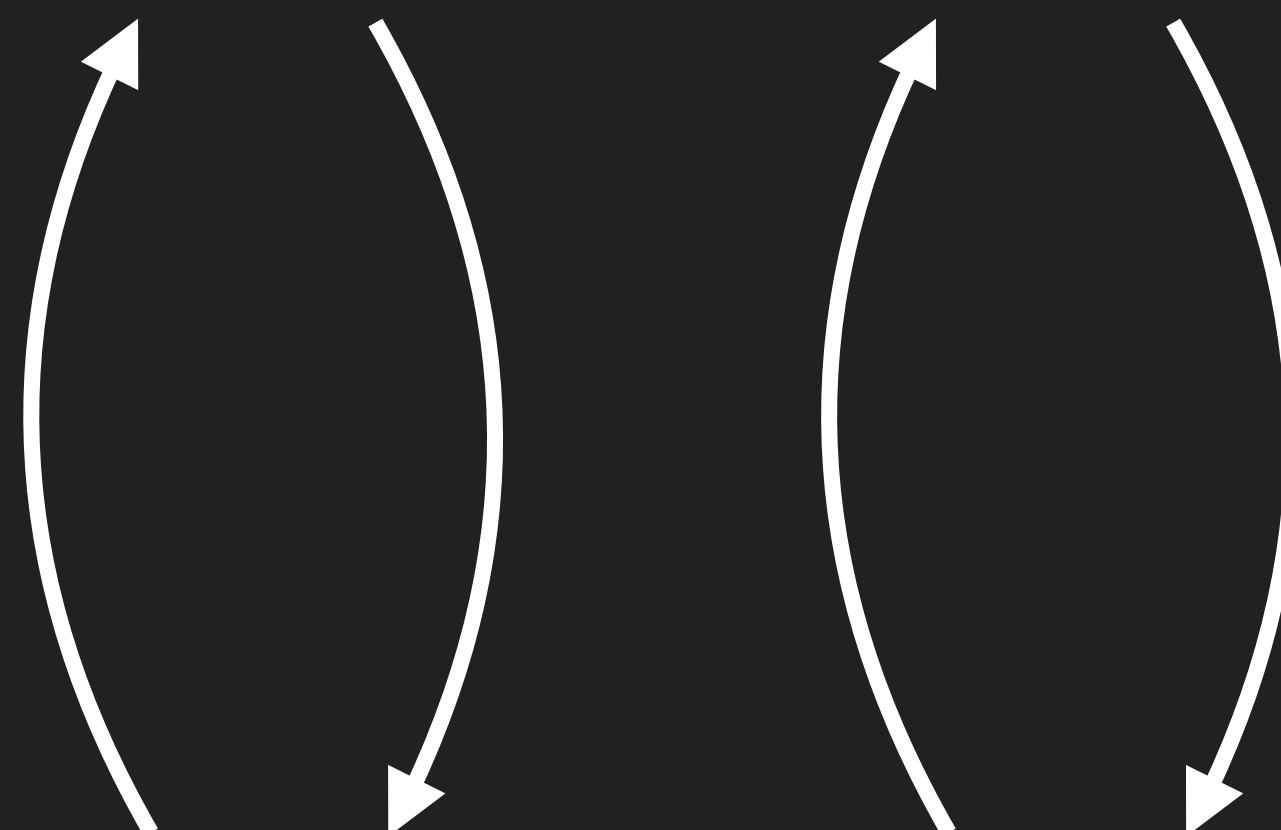


Ceph Storage



16 VCPU
64GB RAM

16 VCPU
64GB RAM



Ceph Storage

If OMERO gets too busy (e.g. >90% load) a new instance is spawned

1. Need to be able to preview image data before accessing it
2. Data preservation when researchers leave lab
3. Prevent unnecessary duplication of image data
4. Facilitate server based analysis pipelines and deter ad hoc manual approaches
5. Reanalysis and sharing of analysis tools



OMERO & BIO-FORMATS:
Extensible Platforms

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Daniel Matthews
drmatthews
Joined on 18 Jun 2012

Popular repositories

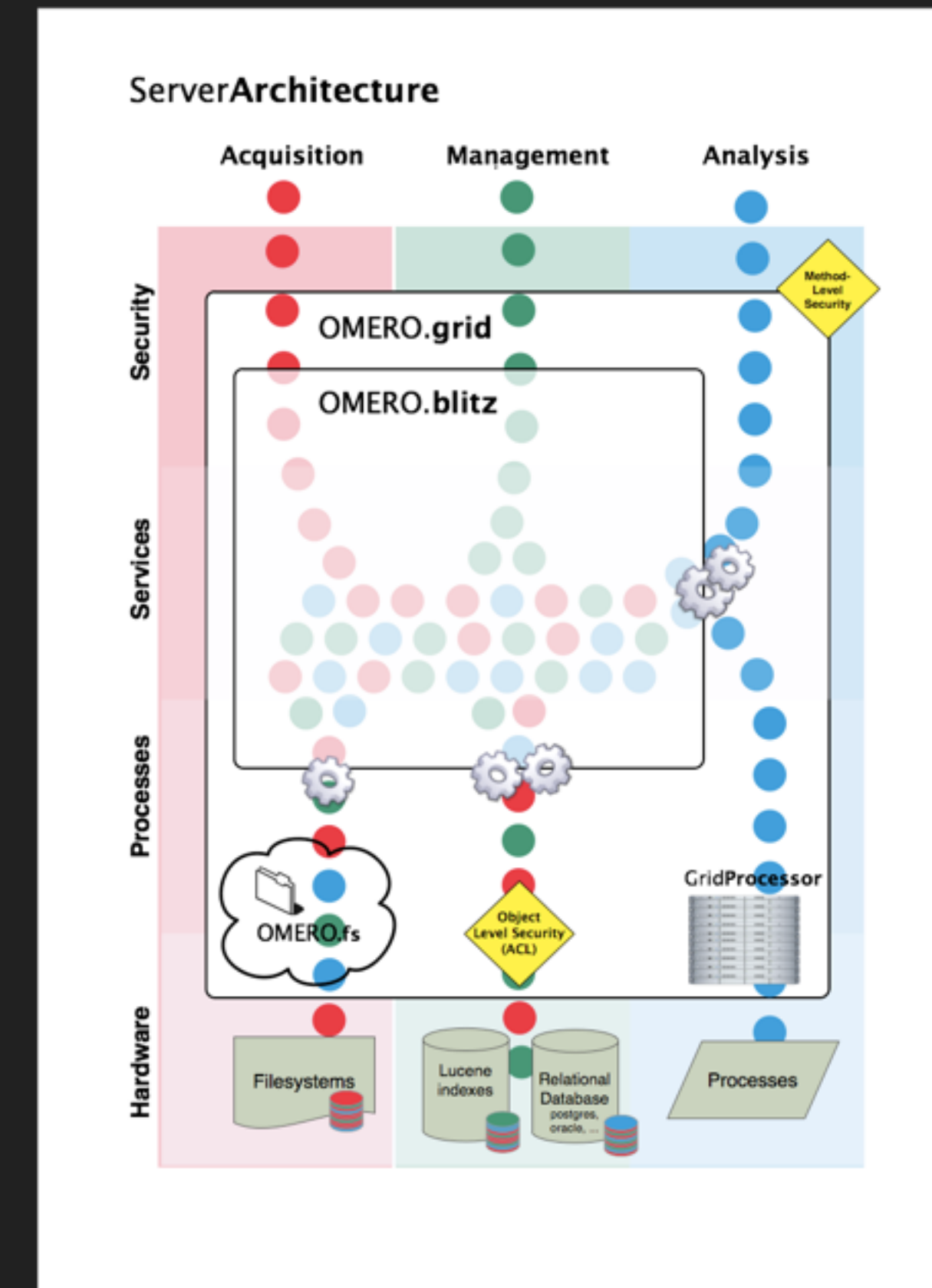
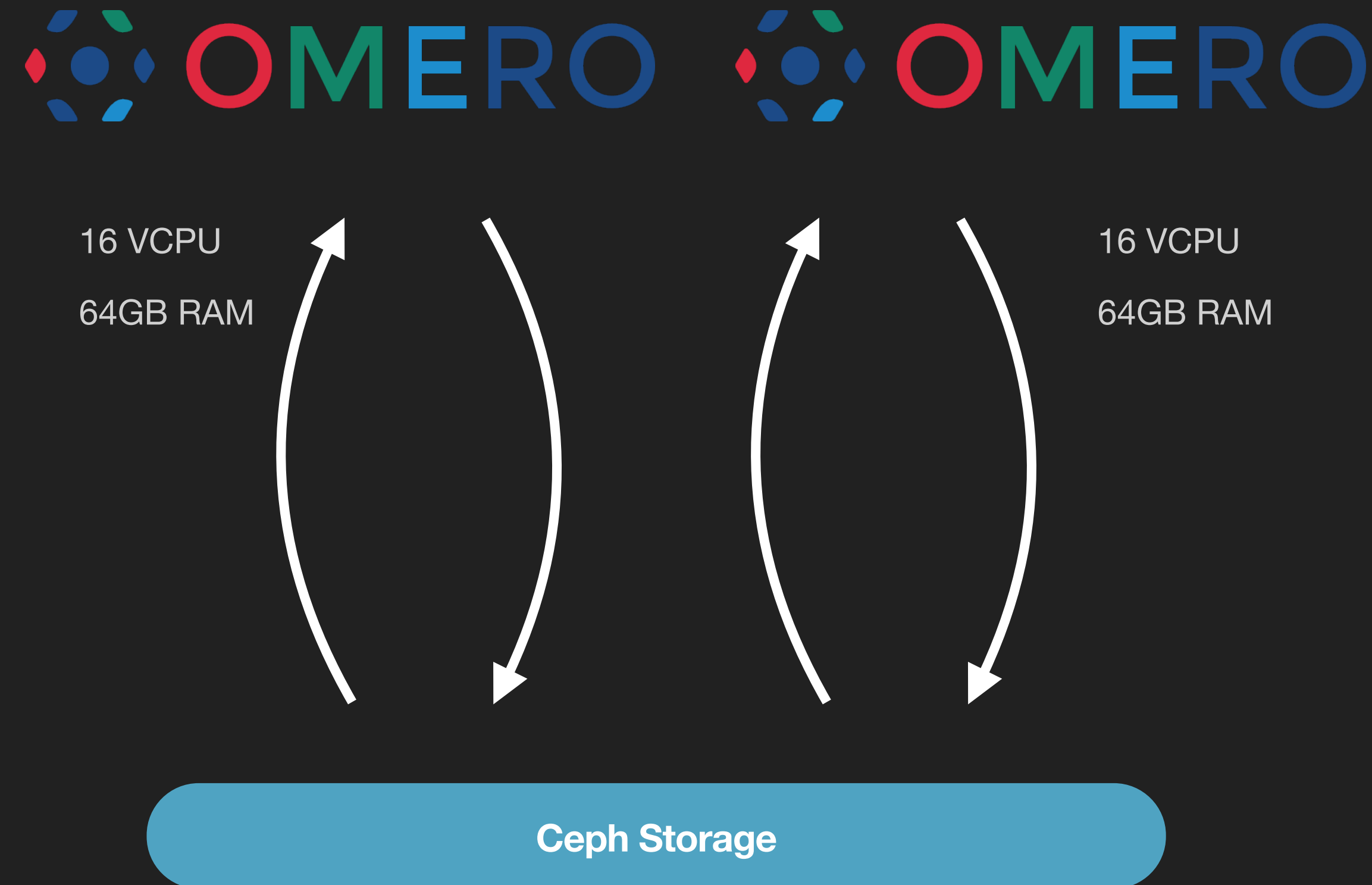
 bioformats Bio-Formats is a Java library for reading and ...	0 ★
 SIMcheck SIMcheck: ImageJ tools for assessing Structur...	0 ★
 CellProfiler CellProfiler is open-source cellular image anal...	0 ★
 simple-STORM Data processing software for dSTORM super-r...	0 ★
 CUDA_SIMrecon Image reconstruction for structured-illuminatio...	0 ★

Repositories contributed to

 QBI-Microscopy/SlideCrop Python app, with wxPython GUI, for automate...	0 ★
 QBI-Microscopy/omero-scripts Utility functions and classes for omero-user-sc...	0 ★
 QBI-Microscopy/Fiji-scripts Jython scripts for Fiji-ImageJ	0 ★
 QBI-Microscopy/Matlab-localisa a collection of scripts for parsing and analysin...	0 ★
 QBI-Microscopy/omero-auto-upl	0 ★

Dan Matthews, QBI

19 



Openstack : Orchestration

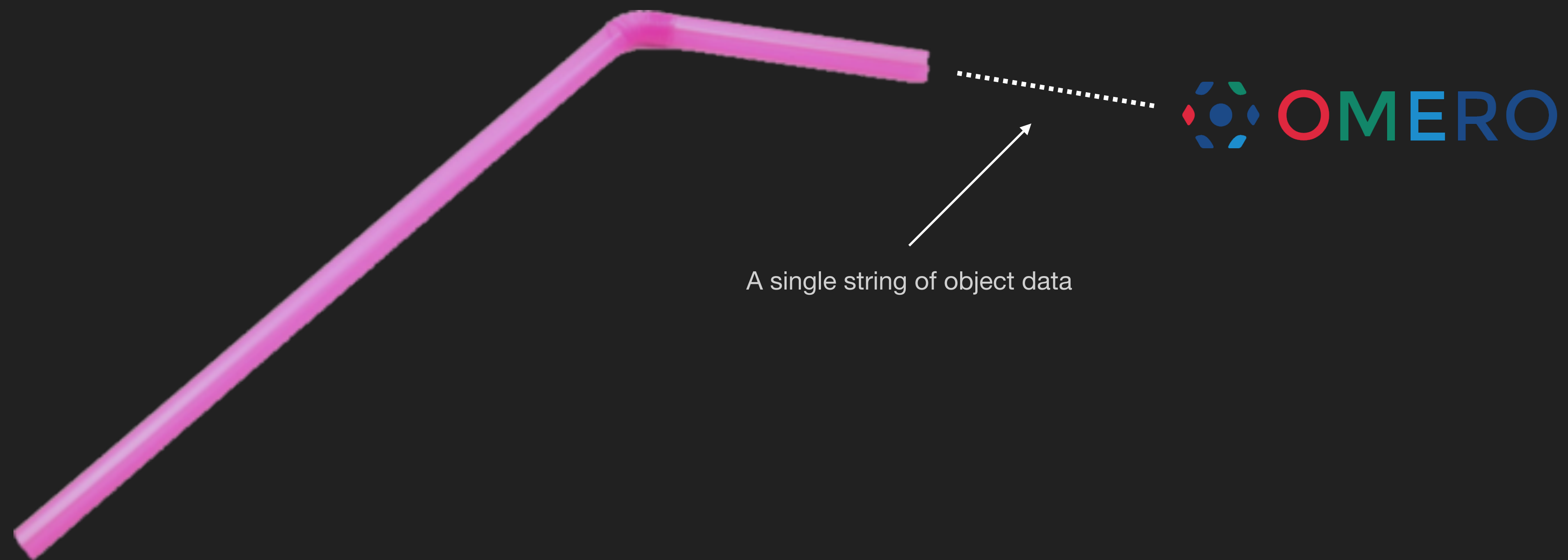
If OMERO gets too busy (e.g. >90% load) a new instance is spawned

BUT new instances are read only to Ceph Storage

The Blitz Gateway



The Blitz Gateway



A single string of object data



* Blitz is also non scalable

Polaris

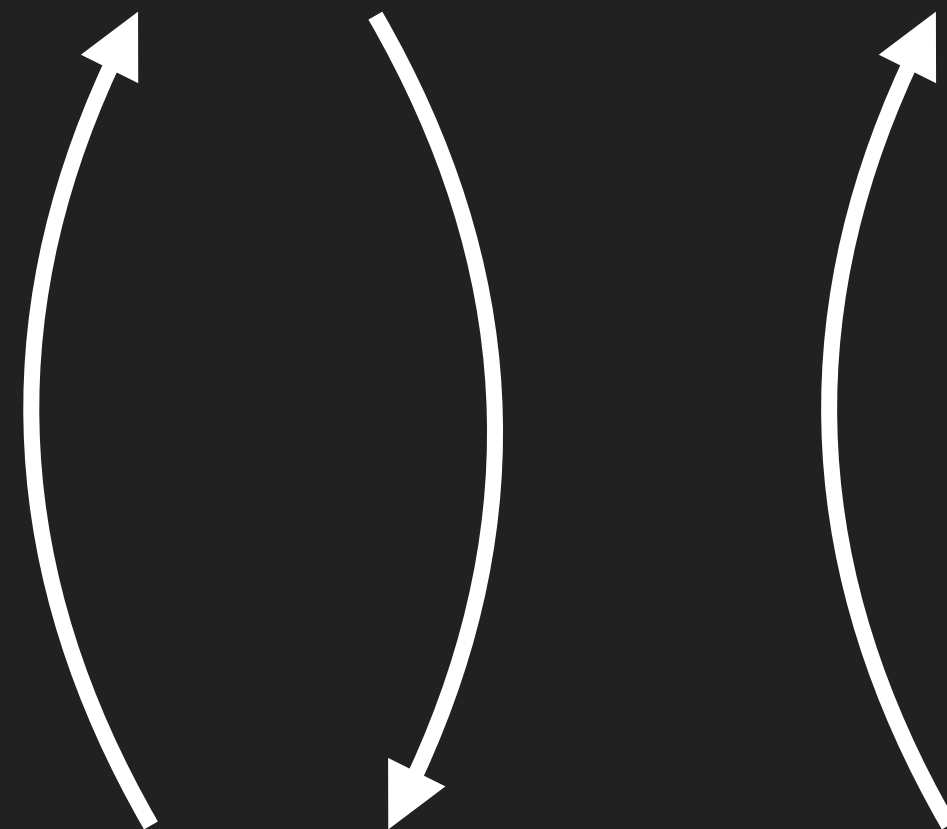
 nectarcloud

 openstack

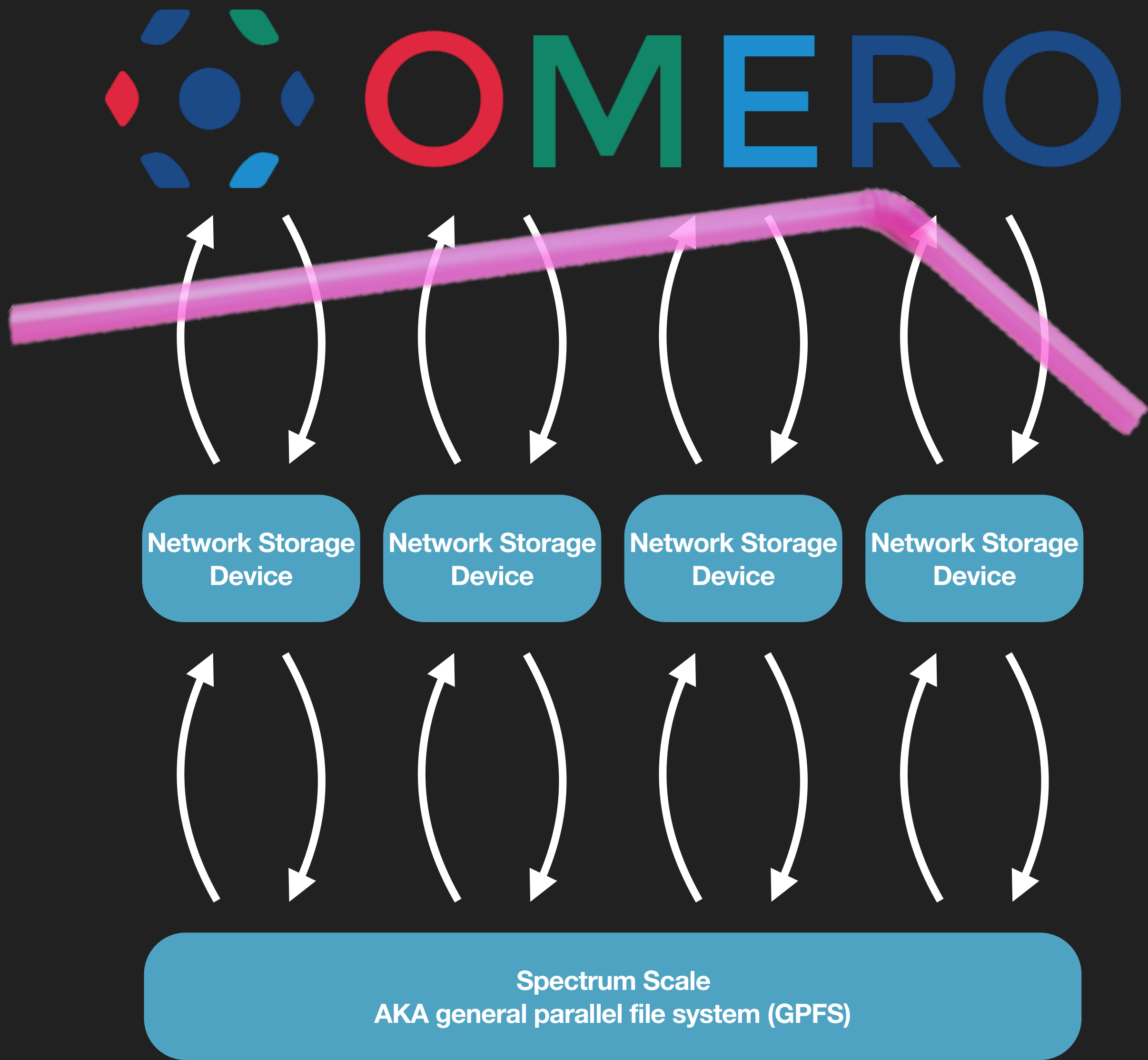
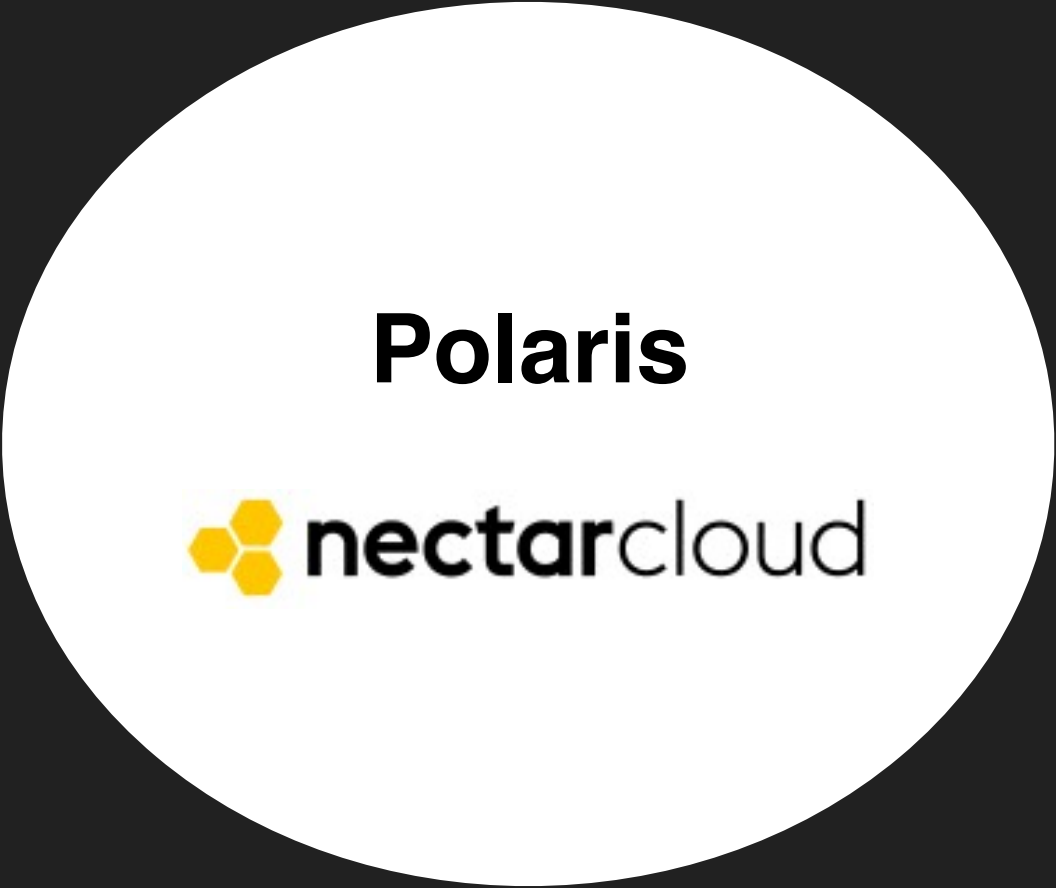
 OMERO  OMERO

16 VCPU
64GB RAM

16 VCPU
64GB RAM




Ceph Storage



16 VCPU
64GB RAM

* no longer requires openstack as a single instance is very fast

The image features a central dark gray rectangular area containing the text "OMERO + big data". This central area is flanked on both the left and right sides by vertical strips of marbled paper. The marbling consists of intricate, swirling patterns of blue, red, orange, and green lines and speckles on a light cream-colored background.

OMERO + big data



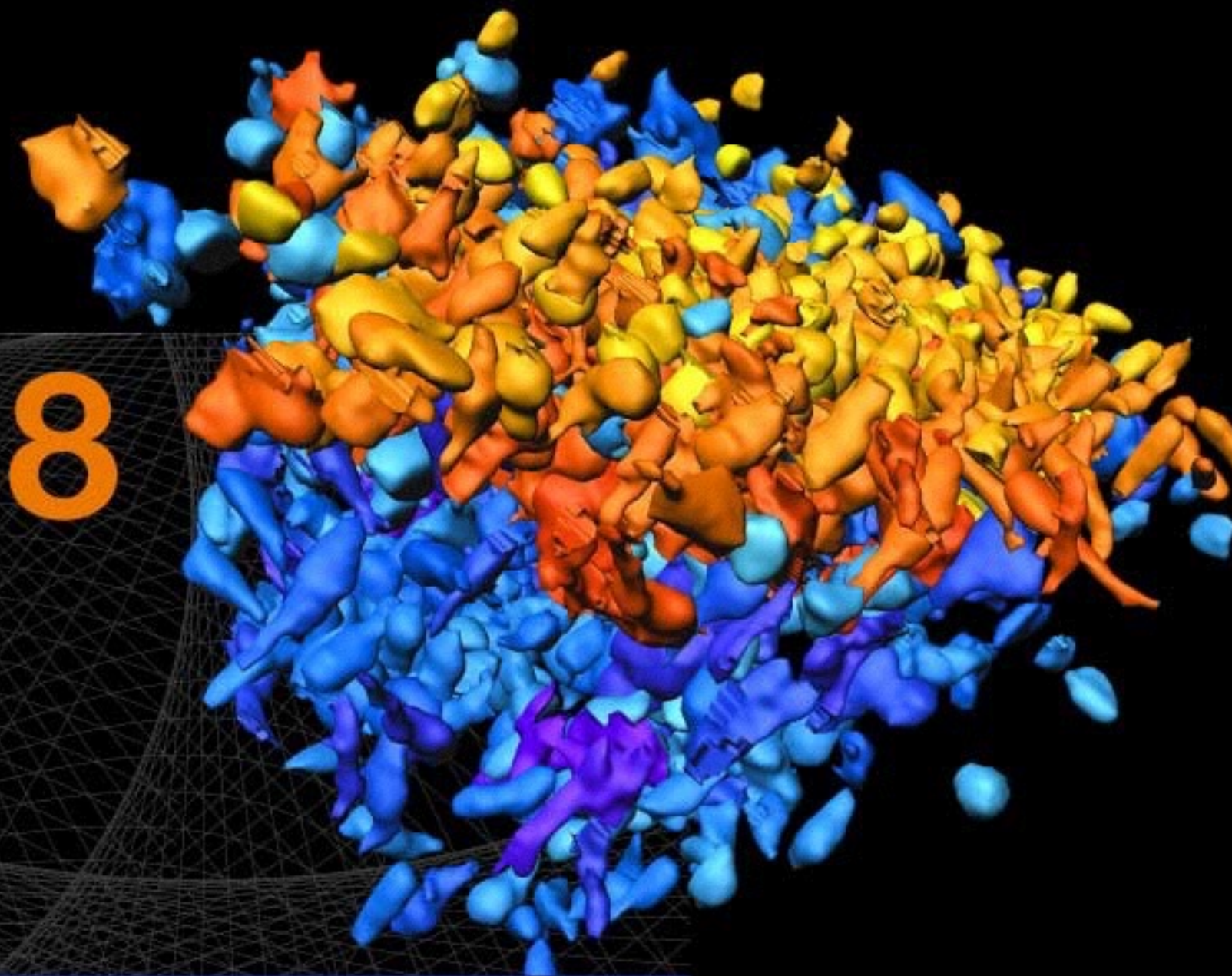
IMARIS8

Setting main frame...

Imaris x64
8.1.2 [Jun 3 2015]
Build 36825 for x64
Copyright © 1993-2015 Bitplane AG
www.bitplane.com
welcome@bitplane.com

BITPLANE
an Oxford Instruments company

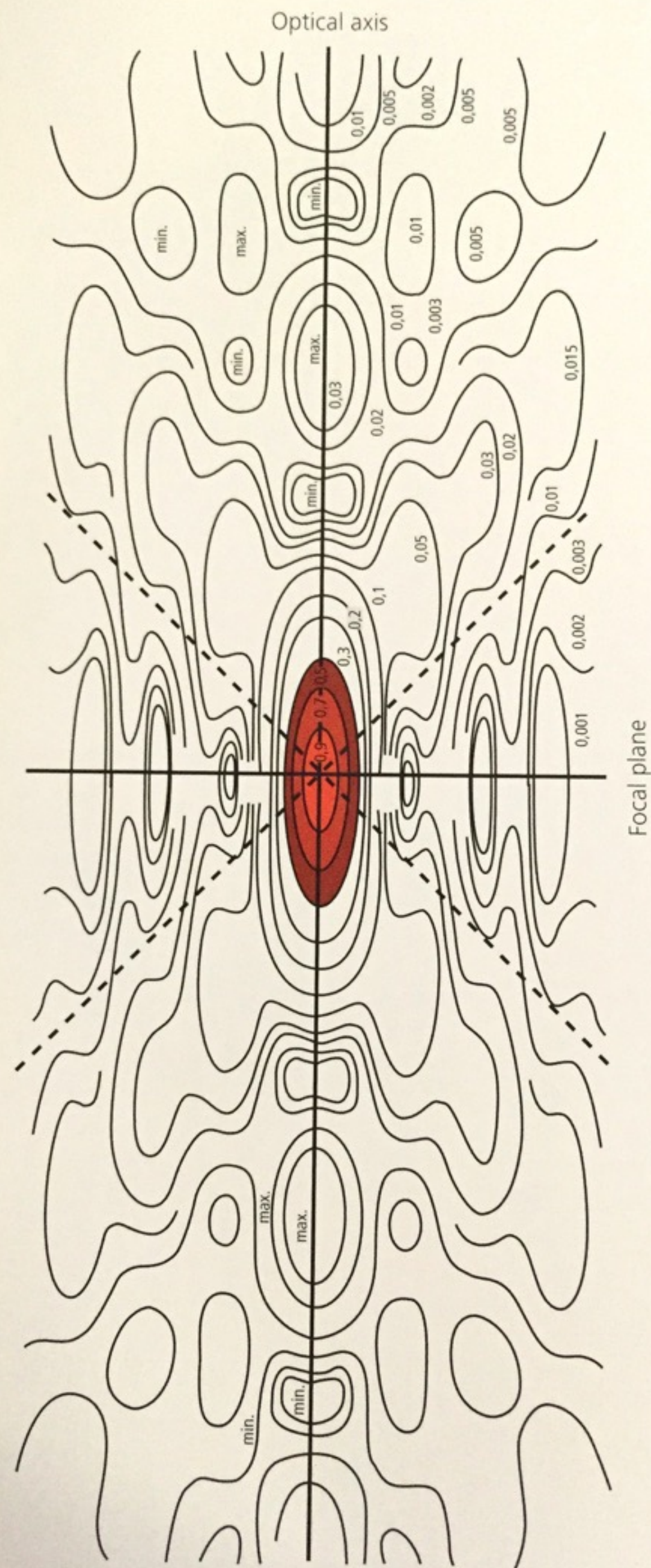
bitplane.com



Deconvolution

Increases image resolution
Increases contrast / SNR
Makes analysis easier / possible

SLOW - 10 - 20 minutes/stack



Huygens GPU Deconvolution server

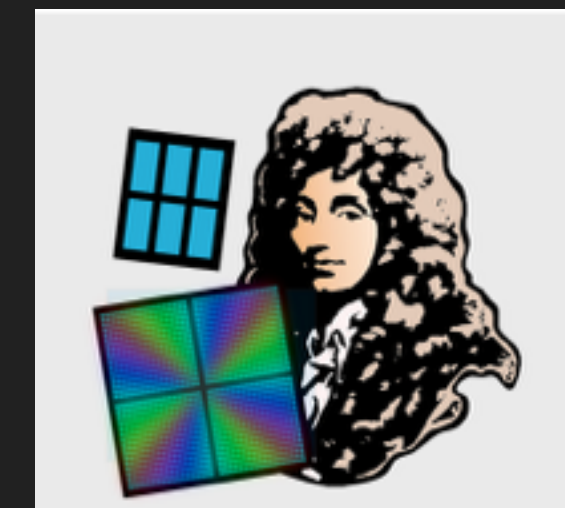
Accessible from anywhere in QBI
Remote and batchable processing

FAST - 20 - 30 seconds / stack

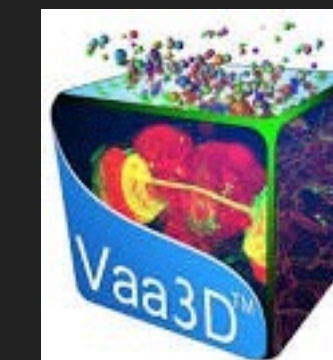
..but teaching researchers to connect
to a remote computer is not always easy



OMERO.Fs



Large nVidia K80 GPU array



work directly with OMERO.Fs and avoid front end / blitz + ICE framework

direct FTP access API or similar to allow queued uploading with correct permissions

back end workflows can make full use of hardware

Essentials required to meet the needs of advanced imaging facilities:

1. Support for large scale images

- Support for large scale images should not be limited. Images 10GB+ are becoming the norm and light-sheet techniques expand this further.
- Data Ingestion:
 - 56432 x 152960px (15.3GB)
 - ~20 mins (6mins to upload, remainder to process into OMERO)
- Extracting ROIs:
 - 77119 x 149376px (7.45GB)
 - 2xpolygon ROIs each (20113 x 31321 px) (1.75 GB)
 - 47 mins
 - 56432 x 152960px (15.3GB)
 - extracting 9xROIs (each 20113 x 31321 px)
 - Session timeout after 6 hrs - not completed

Essentials required to meet the needs of advanced imaging facilities:

1. Support for large scale images

- An exporter client similar to the importer could help to manage this problem and allow the appropriate download procedures to occur in the background.
 - Can we avoid the Blitz gateway?
 - Appears to constrain the handling of binary data and it is assumed that this is where most of the problems in image processing lie
 - We need a parallel mechanism
 - Hadoop FS - works in objects and blocks + add nodes and get more power
- Requires a solution that is built to scale and can drive up hardware utilisation

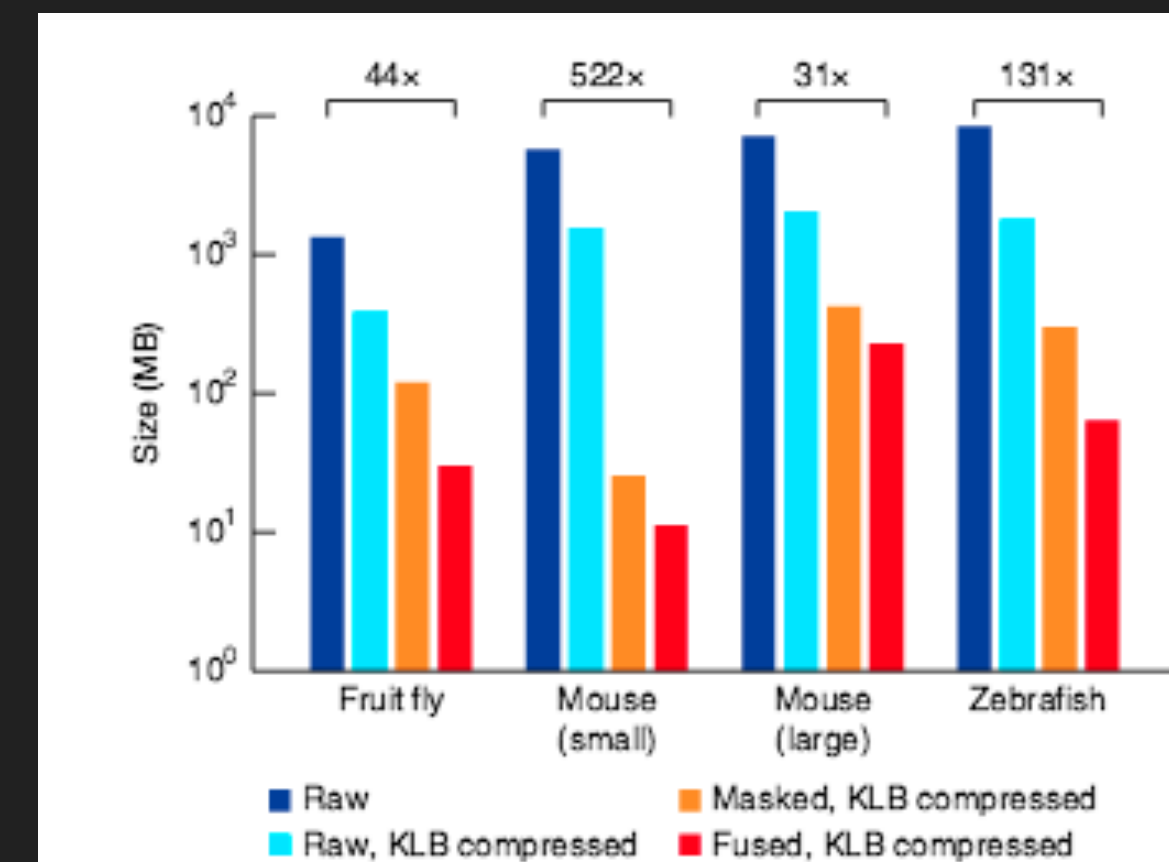
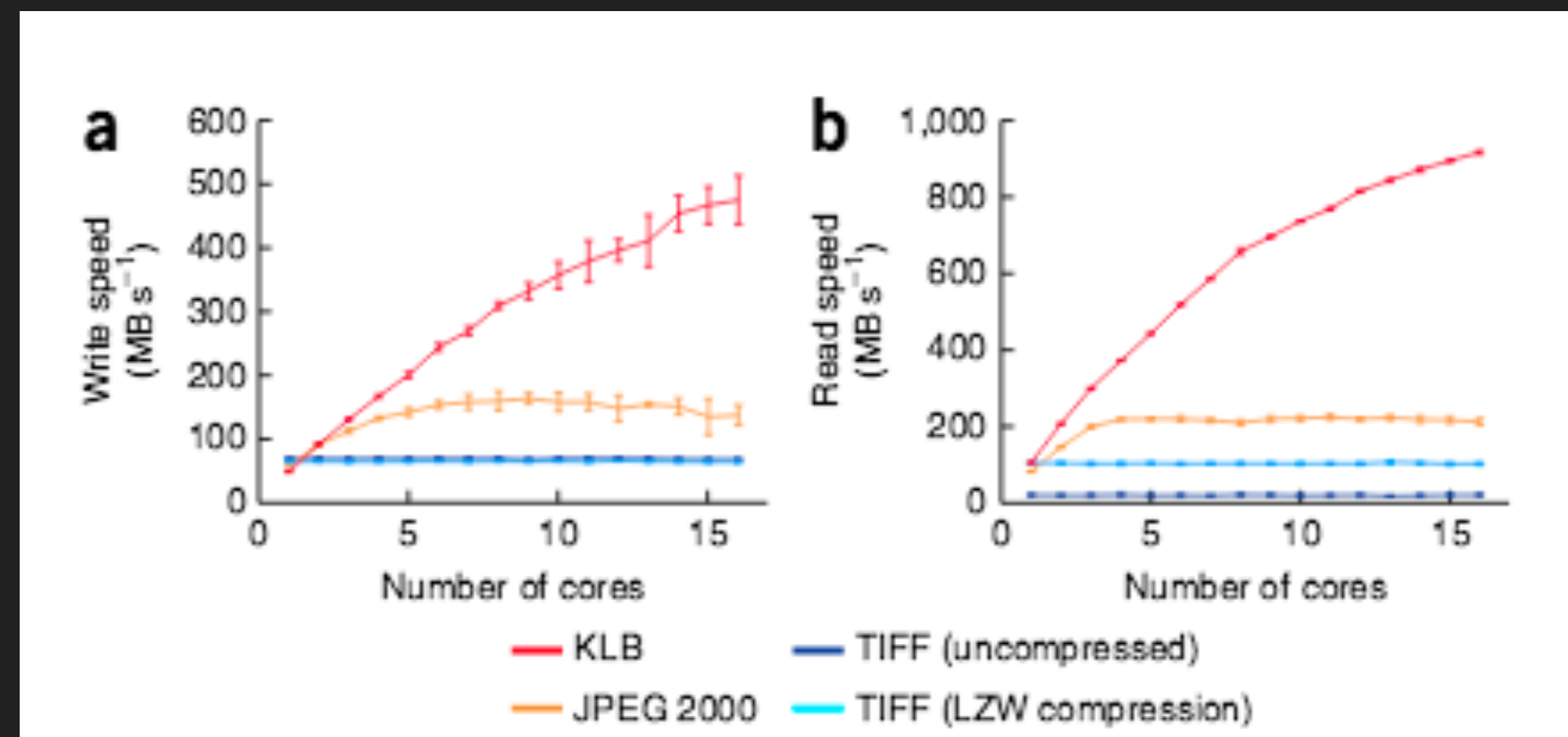
Essentials required to meet the needs of advanced imaging facilities:

- Can we avoid working in raw pixel data?
 - Support for compressed image data rather than having to deal with raw uncompressed pixel data could help speed things up. The time required to manage raw pixel data compared to compressed pixel data can be 10 to 100 fold difference – with datasets of 100's of images, this has a major impact on the user.
 - Attempts to handle ROI extraction in large images by tiling the image has resulted in scripts taking over an hour to complete, beyond the standard timeout of the user session. If the pixel data was compressed, this processing time could be drastically reduced.
 - Attempts to increase the session timeouts has resulted in broken sessions – better integration between the Python and Java API might solve this.

Essentials required to meet the needs of advanced imaging facilities:

- Alternatives to OME TIFF

- TIF and LZW-compressed TIFF = no benefit from multicore CPU architecture
- HDF5 format - data blocks for fast retrieval but slow write (no parallel writing)
- Keller Lab Block (KLB) - data blocks for fast retrieval, parallel write so scales with # of CPU



Efficient processing and analysis of large-scale light-sheet microscopy data

Fernando Amat, Burkhard Höckendorf, Yinan Wan, William C Lemon, Katie McDole & Philipp J Keller

Essentials required to meet the needs of advanced imaging facilities:

2. Support for external processing

- A simple and unhindered means of transferring data in and out of OMERO for external processing (e.g. GPU-cluster analysis)
- One possibility: A direct FTP access API similar to the OMERO Importer to enable queued uploading to OMERO with appropriate permissions and relationships (as these would be large image files, preferably avoiding the restrictions of the ICE framework).



Has the trust of researchers and a great community behind it
but needs scalability and improved hardware utilisation

Microscopy has transitioned from a time of single snapshots to an era of comprehensive imaging
OMERO needs to make this same transition

Parallel file systems help, but only go so far
by liberating OMERO from the blitz gateway imaging research everywhere will become easier and faster

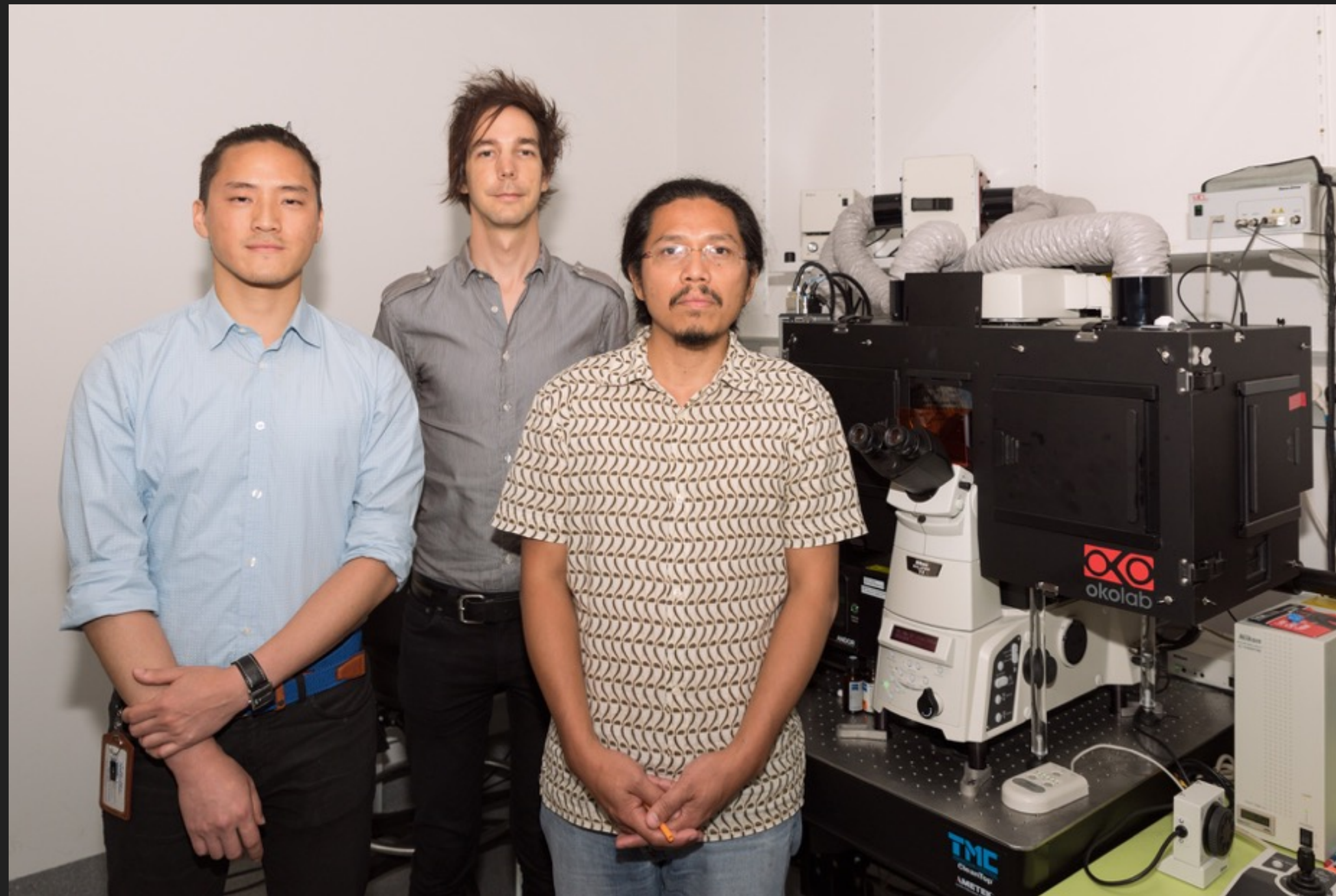
Thank you

Jason Swedlow and the OME team!

Jeremy Ullmann / Boston Children's Hospital

Rumelo Amor and Arthur Chien / QBI Microscopy

Jake Carroll / QBI IT



Jake Carroll

Liz Cooper-Williams
Dan Matthews

Arthur Chien

Rumelo Amor

