Building Bespoke Microscopes & Image Storage

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Bespoke Microscopes

- Why bother?

➡Specific applications

➡Flexibility

➡Cost

Bespoke Microscopes

- Why **NOT** to

➡Cost

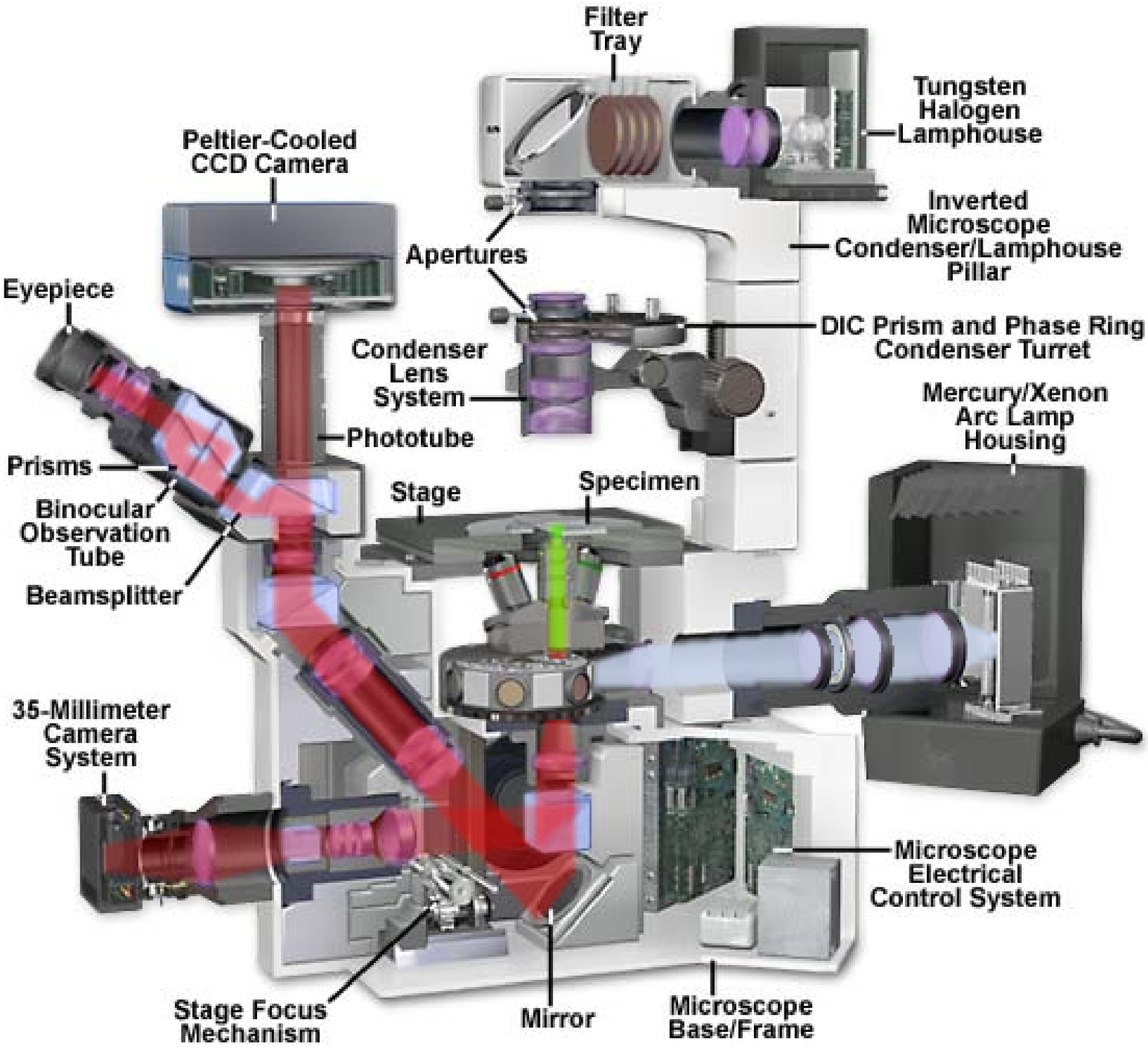
➡Time

➡Usability

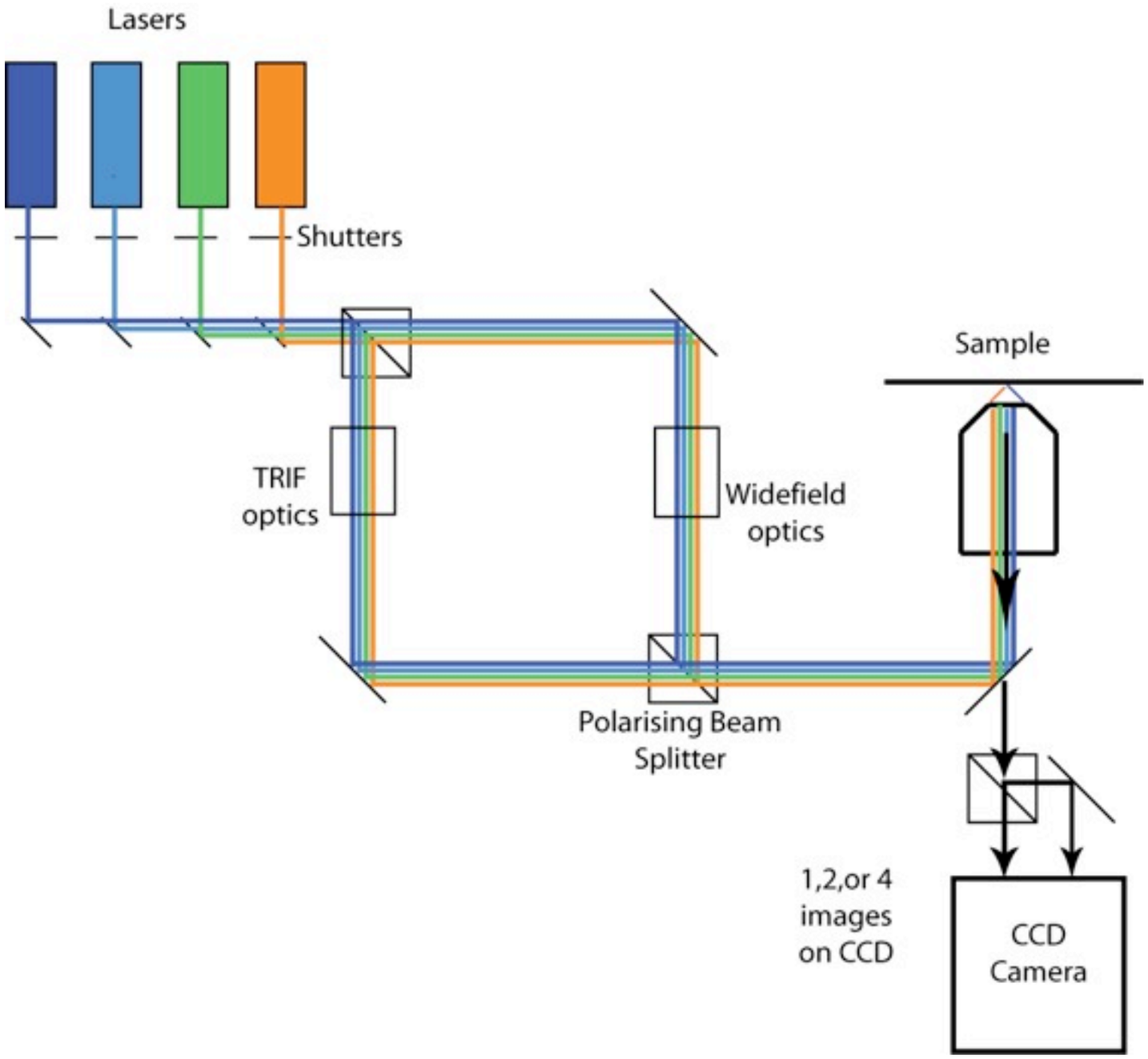
Example Bespoke Microscope

- TIRF - Slimfield Setup
- Built in the biochemistry department in collaboration with Mark Leake (physics).

Conventional microscope



TIRF microscope with split polarisations - schematic

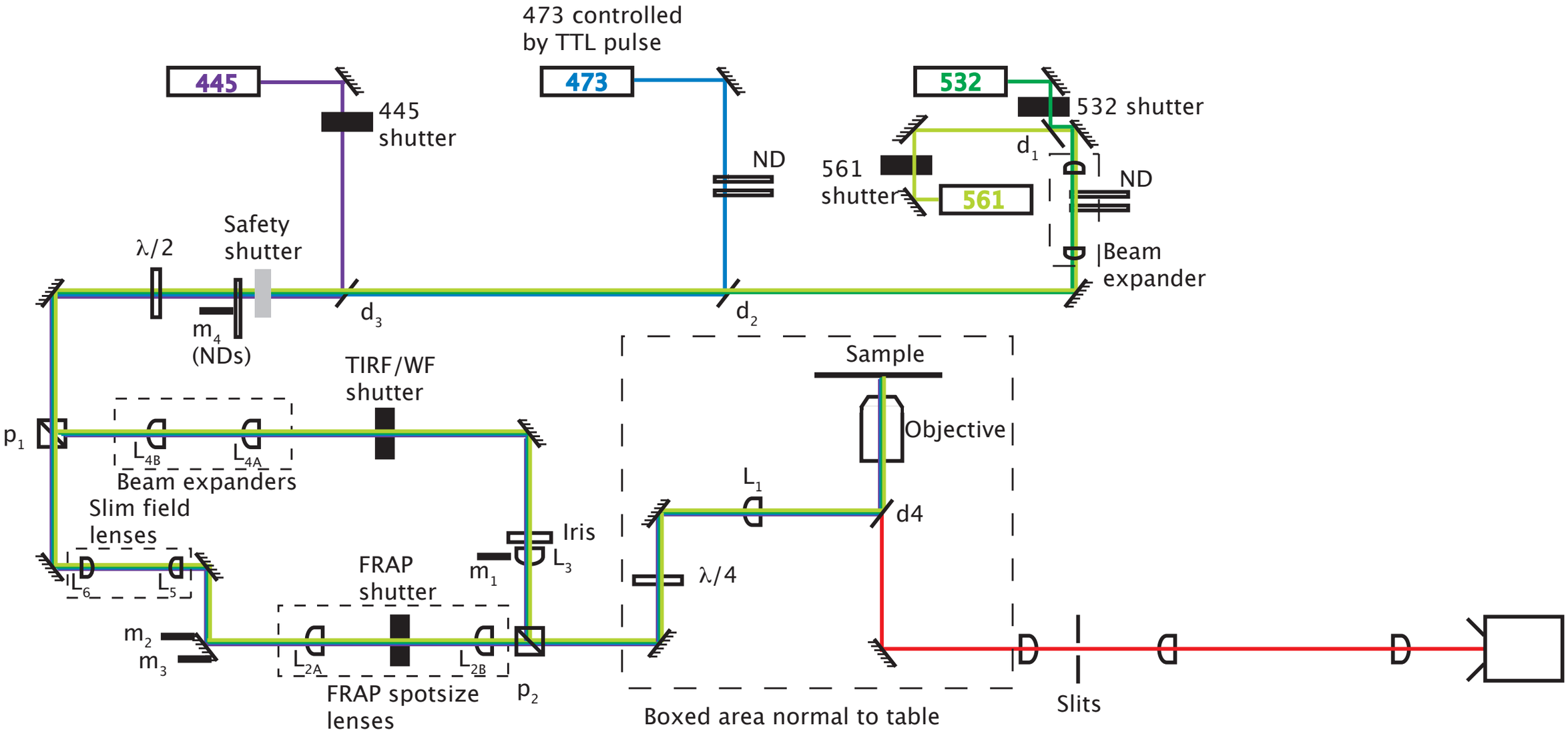


Setup for either

1. TIRF and Widefield
2. Polarisation imaging

Up to 4 images from dual colour in both polarisations

Updated Schematic



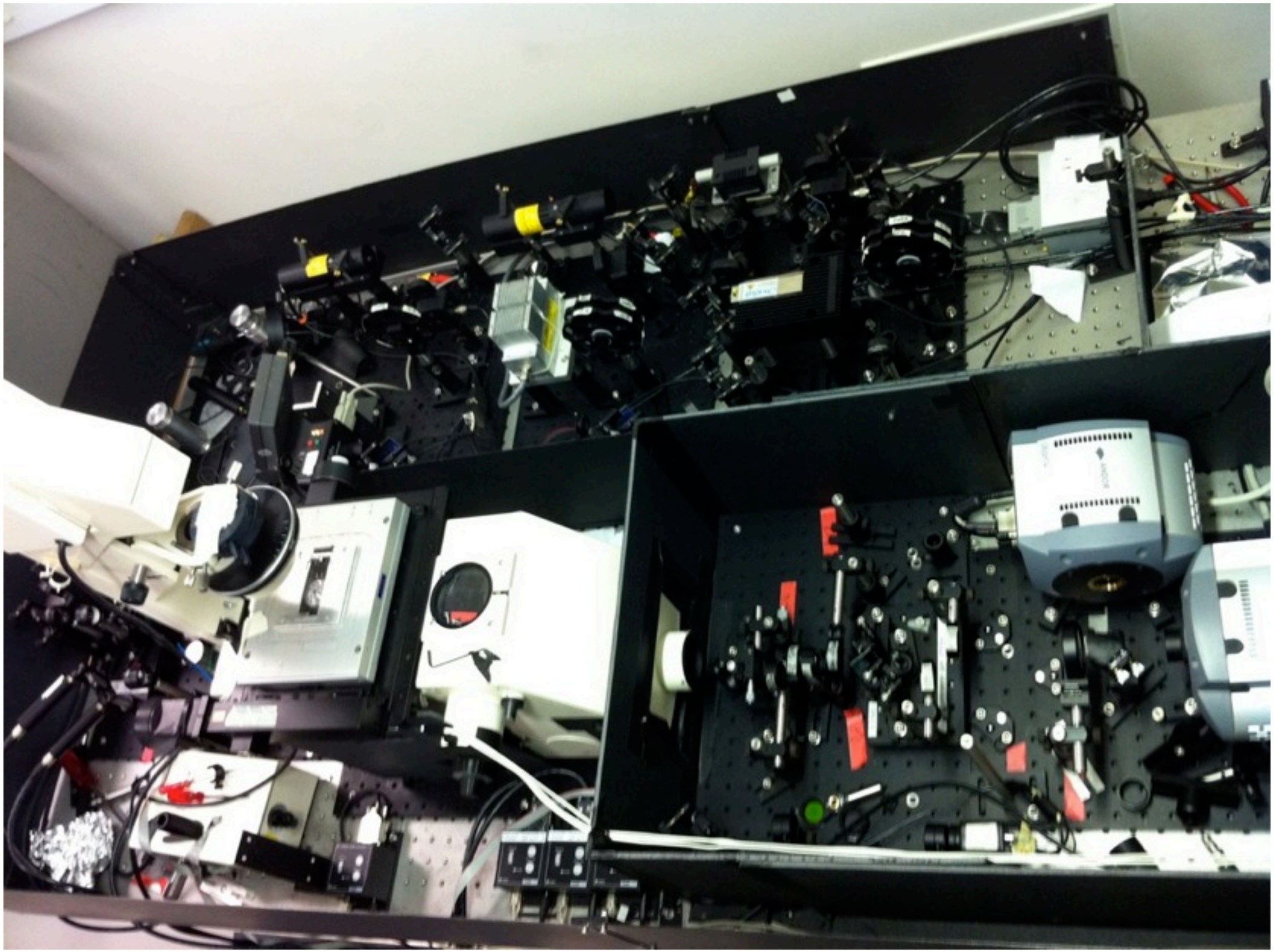
Lenses		Motors	Distances	Dichroics
L_1	f=250 mm	m_1 = TIRF angle	L_1–L_{2A} = 250 + 75	d_1 =
L_{2A}	f=75 mm	m_2 = FRAP spot X	etc....	
L_{2B}	f=200 mm	m_3 = FRAP spot Y		
L_3	f=75?? mm	m_4 = ND wheel		
L_{4A}	f=100?? mm			
L_{4B}	f=200?? mm			
L_5	f=150?? mm			
L_6	f=40?? mm			

Tirf setup

Ian Dobbie 4/1/2009

The real system

The real system II



Features of system

- Multi-colour illumination for widefield, TIRF or slimfield
  - 440, 473, 532, 561
- Dual camera - 512x512 and 128x128
- Simultaneous 2 or 3 colour imaging
- Dual polarisation on excitation and emission
- Dual excitation path for simultaneous photobleaching and imaging.

Advantages of TIRF Slimfield system

- TIRF - slimfield - widefield - FRAP
- More sensitive than commercial system.
- Speed
- Massively more flexible than commercial system.

Disadvantages of TIRF Slimfield system

- Custom written control software.
- Complication.
- Massively more flexible than commercial system.

TIRF-Slimfield system How expensive was it?

- Building costs ~ £100k (hardware)
- Time ~1 person year
- Total cost £150-200k
- Commercial TIRF system ~£150-200k

Should you build a bespoke system?

Yes!

and NO

Justification for Bespoke Systems

- Often necessary for specific specialised problems.
- Easily optimised for several parameters, speed, sensitivity etc...
- Can provide extremely flexible systems

BUT think hard as it is likely to be harder, longer and more expensive than at first thought.

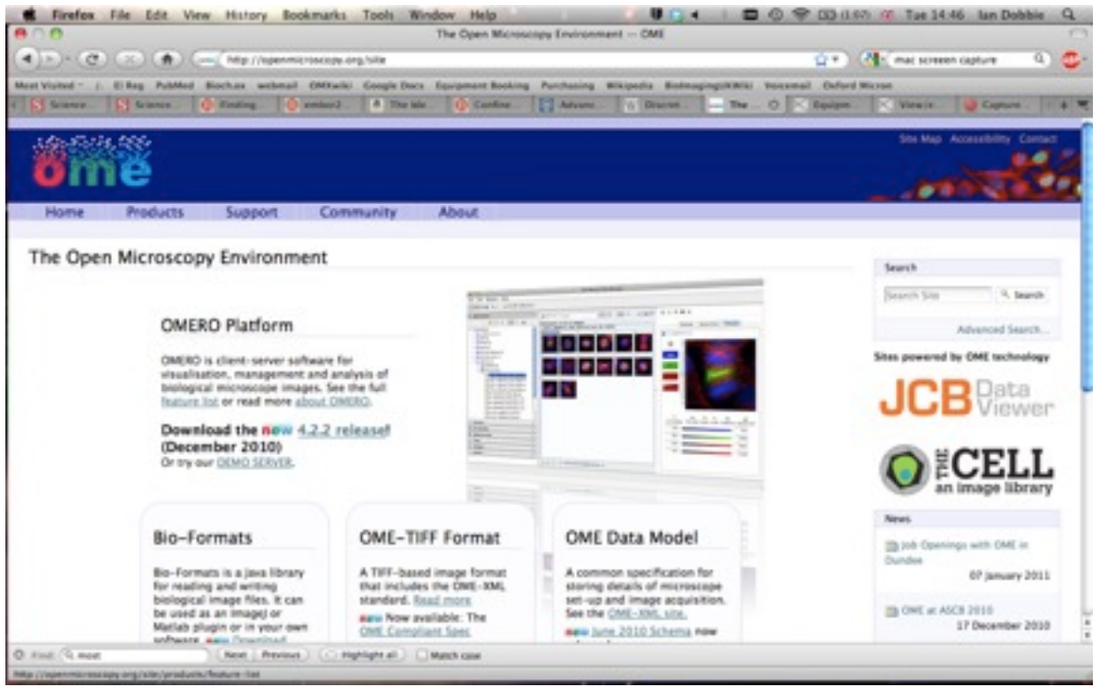
Image Storage

- A major issue especially for 3D, super-resolution and time lapse images
- Where to keep all that data?
- How to organise the data?
- How do you find that data from 3 years ago?

Our Solution - OMERO

- OMERO is a image storage database optimised for microscopy data
- It has a server, where data is stored and several clients that access the data in different ways.

http://openmicroscopy.org/



More reasons for using OMERO

- Allows storage of images from multiple instruments in a single store.
- Metadata is extracted and all images can be labelled with multiple arbitrary tags.
- System is setup so labs can share images, but other users cannot alter your images, just look at them.
- Ensures storage of images for the long term , even when lab members leave.
- You can access your data from anywhere.

Reasons for using OMERO

- Image data is stored on departmental file store: Doesn't count against your quota
- Data is stored in multiple locations and automatically archived to tape, you wont loose it.
- OMERO automatically extracts meta-data and indexes it, allowing rapid searching.

OMERO Clients

Clients are written in Java and run on Mac's, Windows and Linux.

- OMERO.importer - imports images into the database
- OMERO.insight - for everything else.
- OMERO ImageJ plugin - load files from OMERO directly into ImageJ.

OMERO demo

Future plans for OMERO

- Plug image analysis routines into OMERO
- Data will be analysed on the server, so large data sets, or complex analysis can be done.
- Analysis parameters and results will be linked with the original data.

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