QUAREP-LiMi

QUAREP-LiMi: a unified approach to Microscope Quality Control

Glyn Nelson

Newcastle University

The BioImaging Unit

Newcastle University

Why perform Quality Control?

- Maintenance of advanced fluorescent microscopes is essential to allow researchers to have full confidence in the imaging data collected
    - Observations made should be the result of the observed biology not changes in microscope performance
    - Within the image and over time

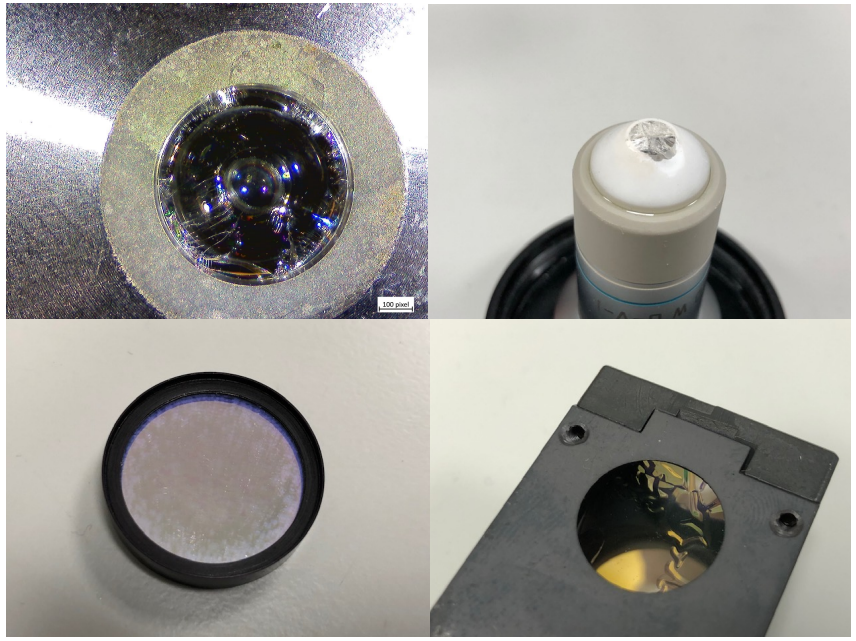
- Microscope-based imaging is becoming more quantitative

- All microscope systems degrade or change over time
    - Filters can become burned
    - Light source characteristics change
    - Detector sensitivity may reduce
    - Damage

- The quality of any observation and imaging data can only be as good as the quality microscope used to make them: thus, it is important that microscope *'quality'* is understood and documented to support intensity and localisation data

- Microscope systems are expensive
    - Make best use of the investment

Quality Control: why bother…..?



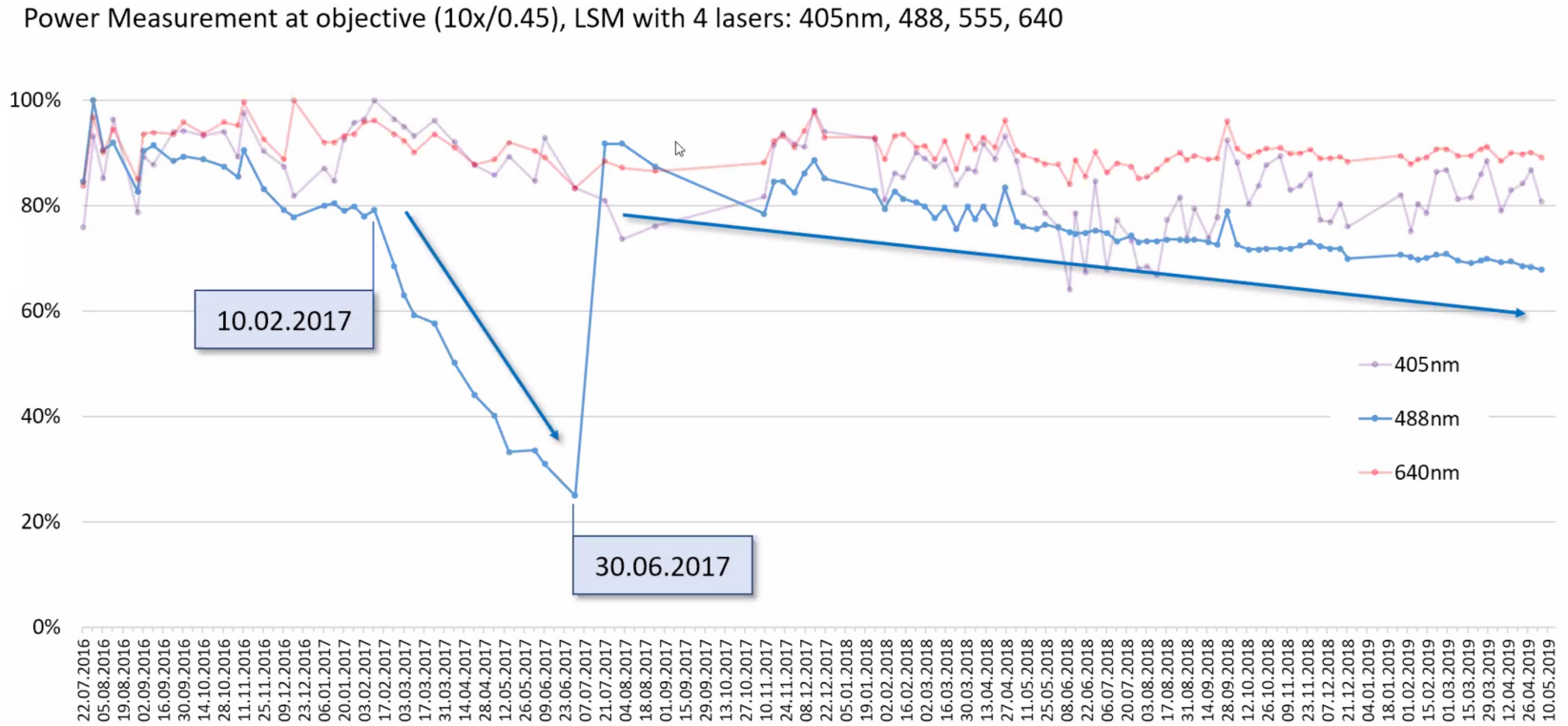
- 80% of reported ‘quality issues’ to facility staff are from the sample or lens
- Clean the surface of the coverslip – free from old oil, mountant or dried media?
- Clean lenses before and after use and remove any old oil
- Has oil been placed on an air objective…?

Example tracking laser power: if you don't measure, you don't know.

Fluorescence microscopy is now being used as a 'quantitative' tool to estimate protein abundance in tissues and cells

Understanding the performance of the illumination and detector systems is important.

-detector systems more stable than the illumination

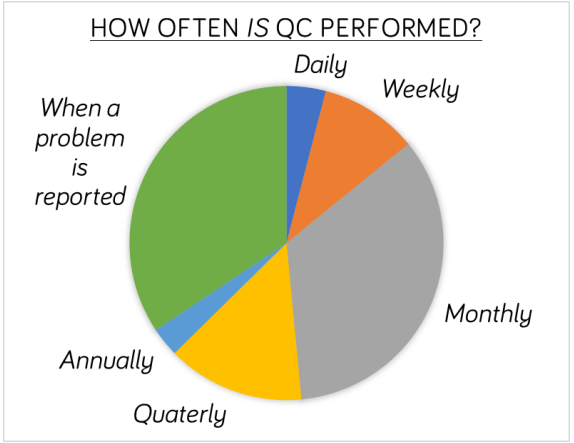


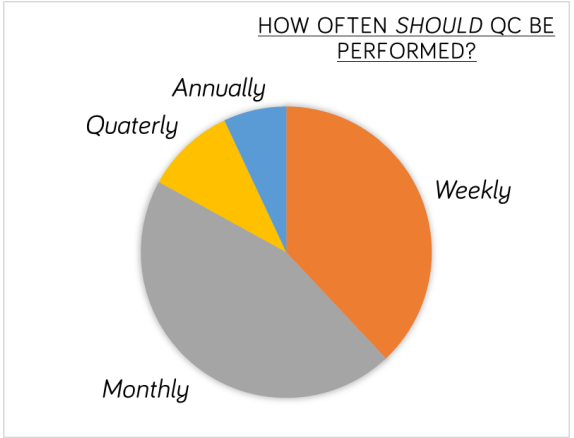
Laurent Gelman, QUAREP WG1

Quality Control: the problem

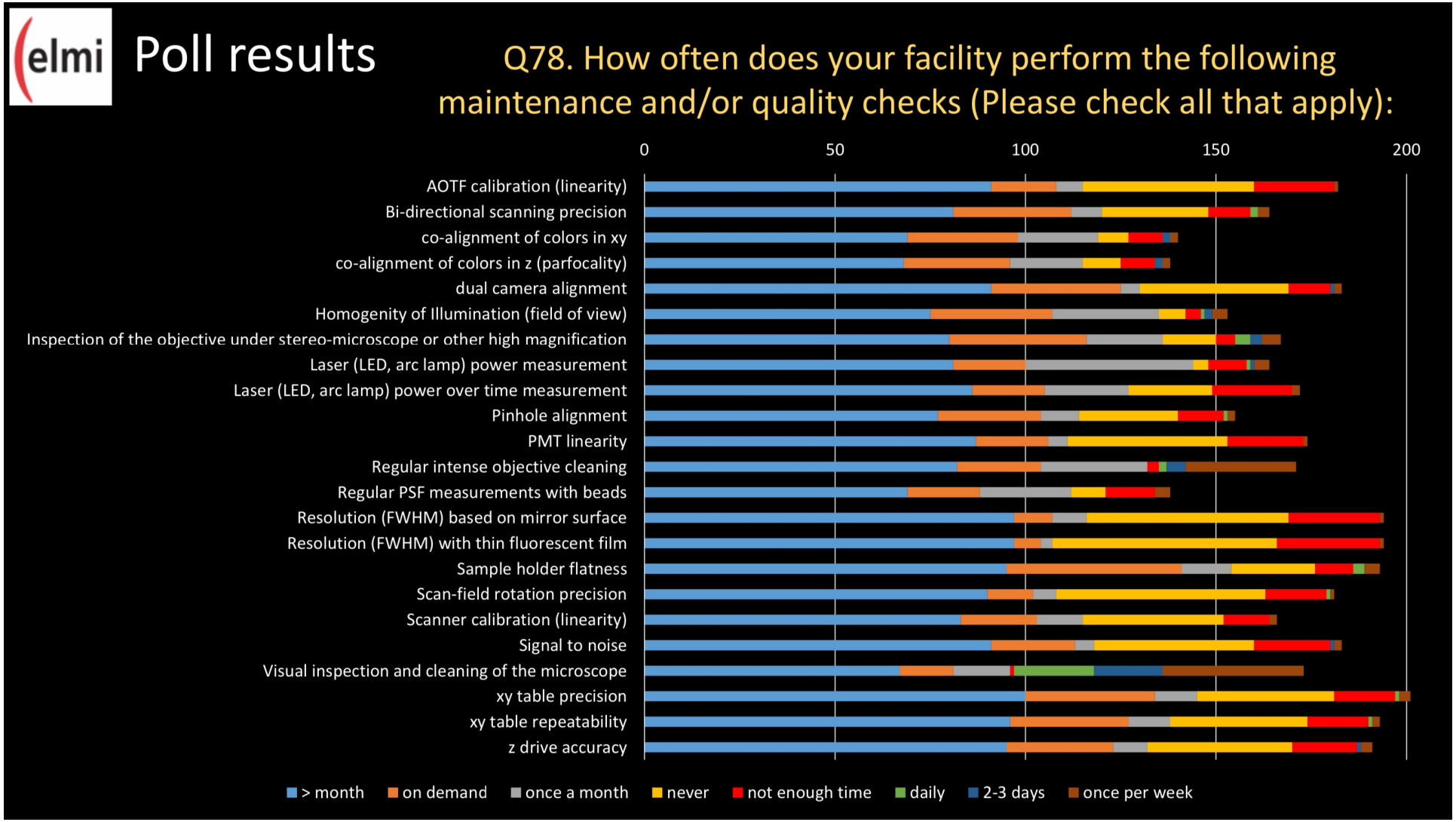
- Not all microscope custodians are ‘microscopists’
  - Experience and time will dictate what tests are performed and how often
- No currently accepted community standards for microscope and image ‘quality’
  - What standards?
  - What tests?
  - How to perform them?
  - How often?
- Time consuming
  - Dictated by available time – reactive rather than proactive
  - Make way for higher priority activities
- Little manufacturer input
  - System performance upon installation
  - Accepted tolerances
  - Inbuilt QC tests
- Journals expect raw data to be available upon request (inc metadata)
  - Nothing in the metadata describes microscope performance

“Microscope QC practices within the UK microscopy facilities” survey 2019



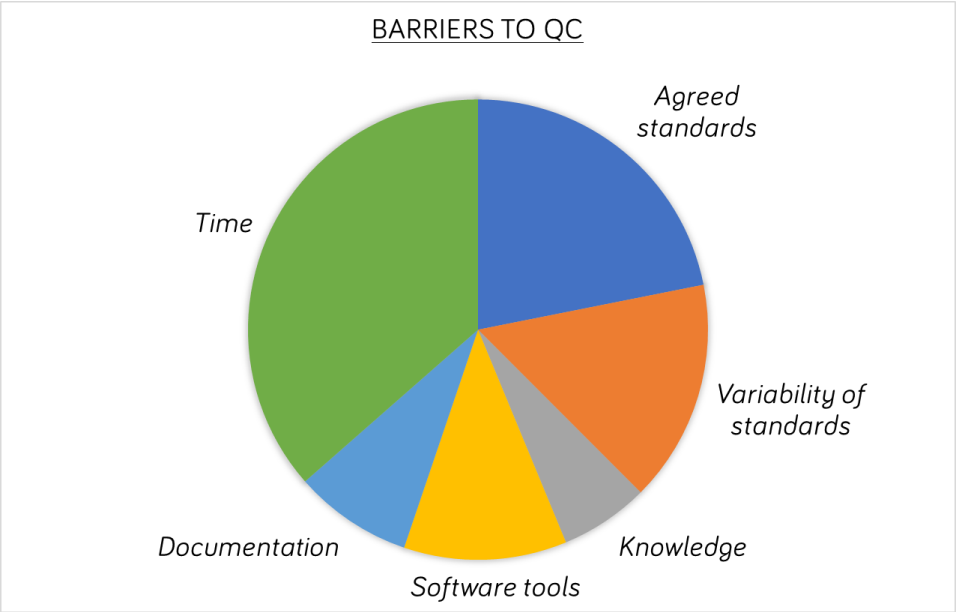


Quality Control: Frequency Performed



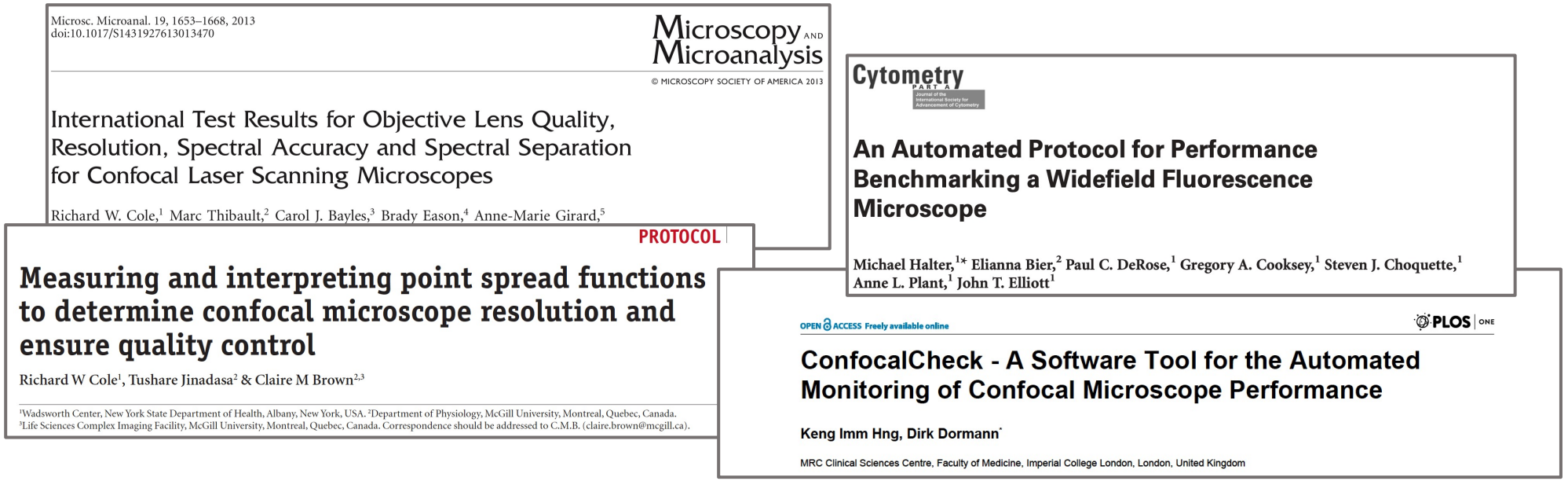
"Microscope QC practices" global core facility & microscope user survey 2020

Quality Control: Barriers to Performance



"Microscope QC practices within the UK microscopy facilities" survey 2019

Quality Control: why bother…..?



Microsc. Microanal. 19, 1653–1668, 2013
doi:10.1017/S1431927613013470

Microscopy AND Microanalysis

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International Test Results for Objective Lens Quality, Resolution, Spectral Accuracy and Spectral Separation for Confocal Laser Scanning Microscopes

Richard W. Cole,[1] Marc Thibault,[2] Carol J. Bayles,[3] Brady Eason,[4] Anne-Marie Girard,[5]

Cytometry PART A
Journal of the International Society for Advancement of Cytometry

An Automated Protocol for Performance Benchmarking a Widefield Fluorescence Microscope

Michael Halter,[1]* Elianna Bier,[2] Paul C. DeRose,[1] Gregory A. Cooksey,[1] Steven J. Choquette,[1] Anne L. Plant,[1] John T. Elliott[1]

PROTOCOL

Measuring and interpreting point spread functions to determine confocal microscope resolution and ensure quality control

Richard W Cole[1], Tushare Jinadasa[2] & Claire M Brown[2,3]

[1]Wadsworth Center, New York State Department of Health, Albany, New York, USA. [2]Department of Physiology, McGill University, Montreal, Quebec, Canada. [3]Life Sciences Complex Imaging Facility, McGill University, Montreal, Quebec, Canada. Correspondence should be addressed to C.M.B. (claire.brown@mcgill.ca).

OPEN ACCESS Freely available online

PLOS ONE

ConfocalCheck - A Software Tool for the Automated Monitoring of Confocal Microscope Performance

Keng Imm Hng, Dirk Dormann*

MRC Clinical Sciences Centre, Faculty of Medicine, Imperial College London, London, United Kingdom

"Nice to have, but sample and biological variance make this unnecessary "

reviewer's comment to BBSRC TDRF funding application 2016

"…. Quality control should be driven by manufacturers not users of instruments"

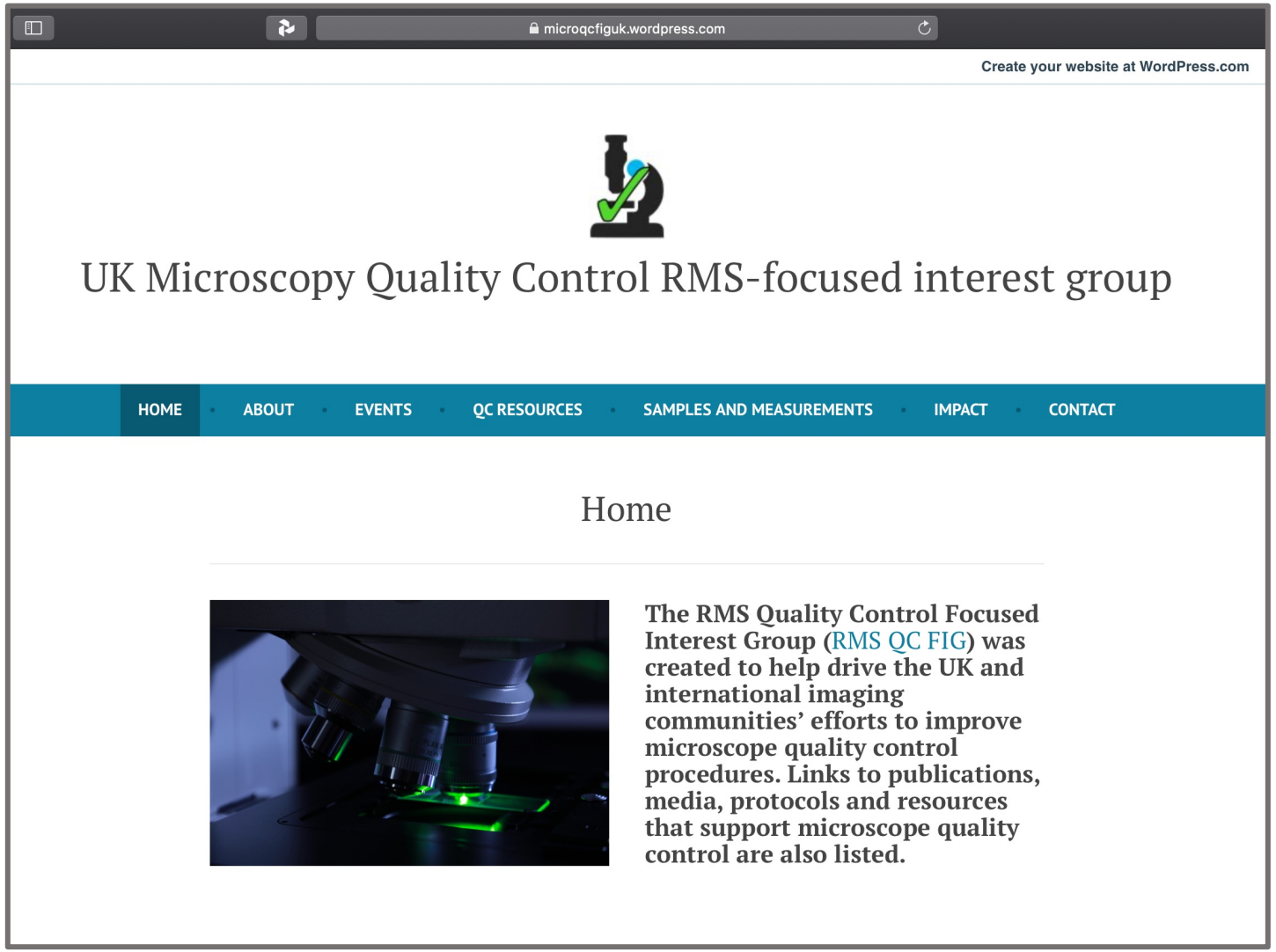
review panel comment on WT Technology development funding application 2020

RMS driven UK based QC group

https://www.rms.org.uk/network-collaborate/focussed-interest-groups/quality-control.html

https://microqcfig.org

Royal Microscopical Society



QUAREP-LiMi

INTERNATIONAL STANDARD

ISO 21073

First edition
2019-12

Microscopes — Confocal microscopes — Optical data of fluorescence confocal microscopes for biological imaging

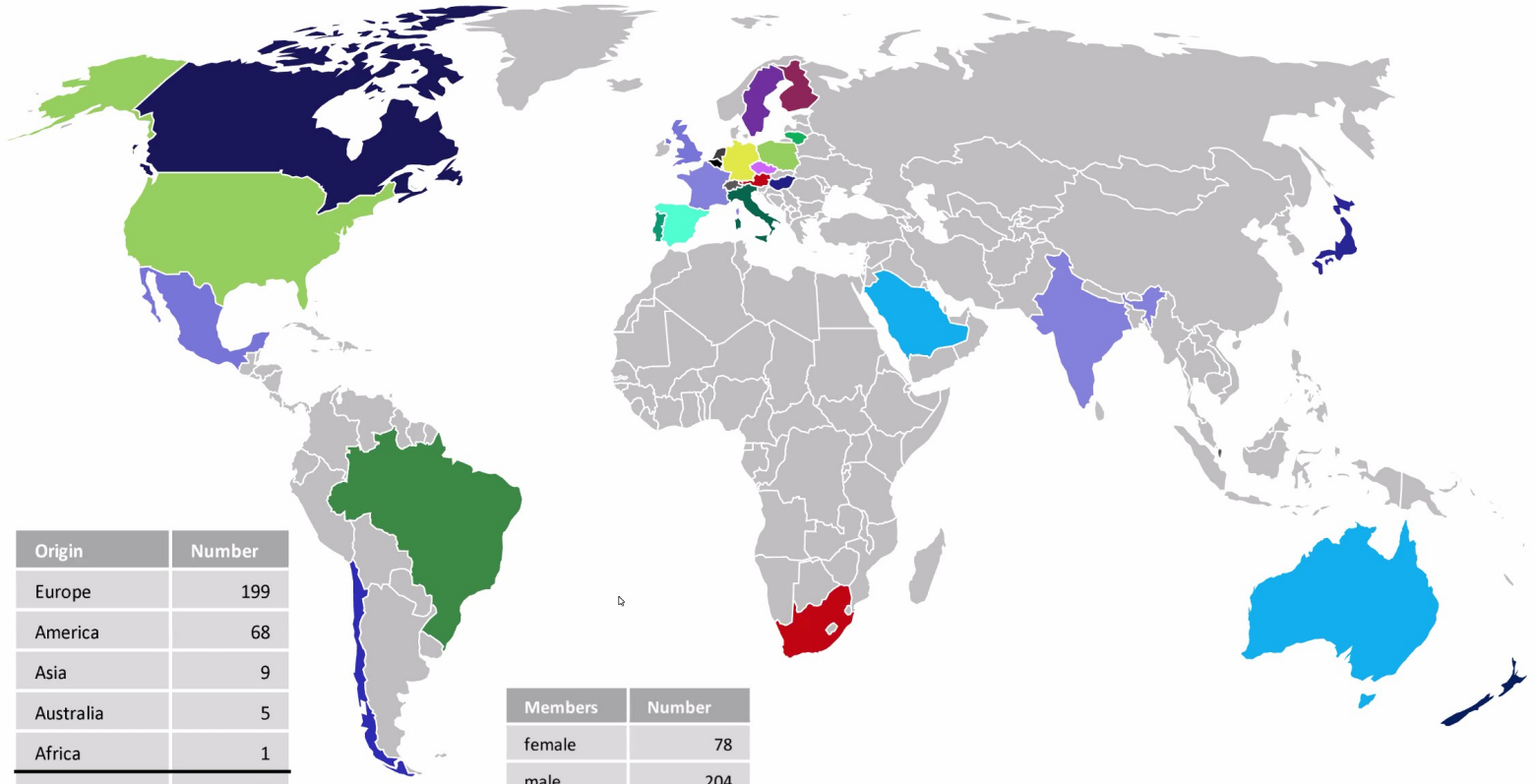
Microscopes — Microscopes confocaux — Données optiques des microscopes confocaux à fluorescence pour l'imagerie biologique

Nelson, G., Gelman, L., Faklaris, O., Nitschke, R. & **Laude, A**. Interpretation of confocal iso 21073: 2019 confocal 628 microscopes: Optical data of fluorescence confocal microscopes for biological imaging- recommended methodology for quality control. arXiv DOI: https://arxiv.org/abs/2011.08713 (2020).

QUAREP-LiMi - Where do we come from and who are we?

QUAREP-LiMi

Origin	Number
Australia	4
Austria	7
Belgium	3
Canada	12
Czech Republic	2
Chile	2
Finland	10
France	21
Germany	89
Great Britain	32
Hungary	1
India	1
Italy	3
Japan	4
Lithuania	1
Mexico	1
Netherlands	7
New Zealand	1
Poland	1
Portugal	6
Saudi Arabia	1
Singapore	3
South Africa	1
Spain	2
Sweden	4
Switzerland	12
United States	51



Origin	Number
Europe	199
America	68
Asia	9
Australia	5
Africa	1
Total	282

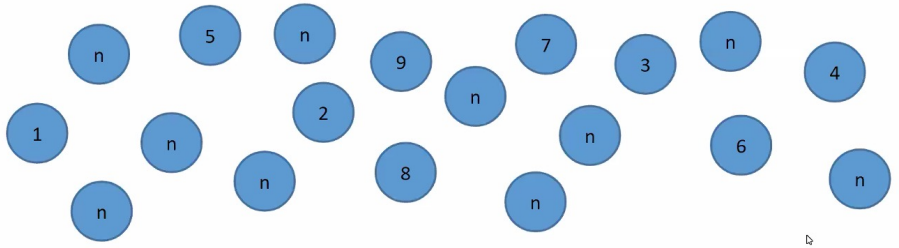
Members	Number
female	78
male	204
Total	282

World map by www.freeworldmaps.net

www.miap.eu Ni 06-2021 Status: 06/20/2021

QUAREP-LiMi - Who are we?

Microscopy Community



Affiliation	Number
Academia	188
Industry	72
Standards Org.	13
Others	9
Total	282

05/21/2021

FOCUS | COMMENT

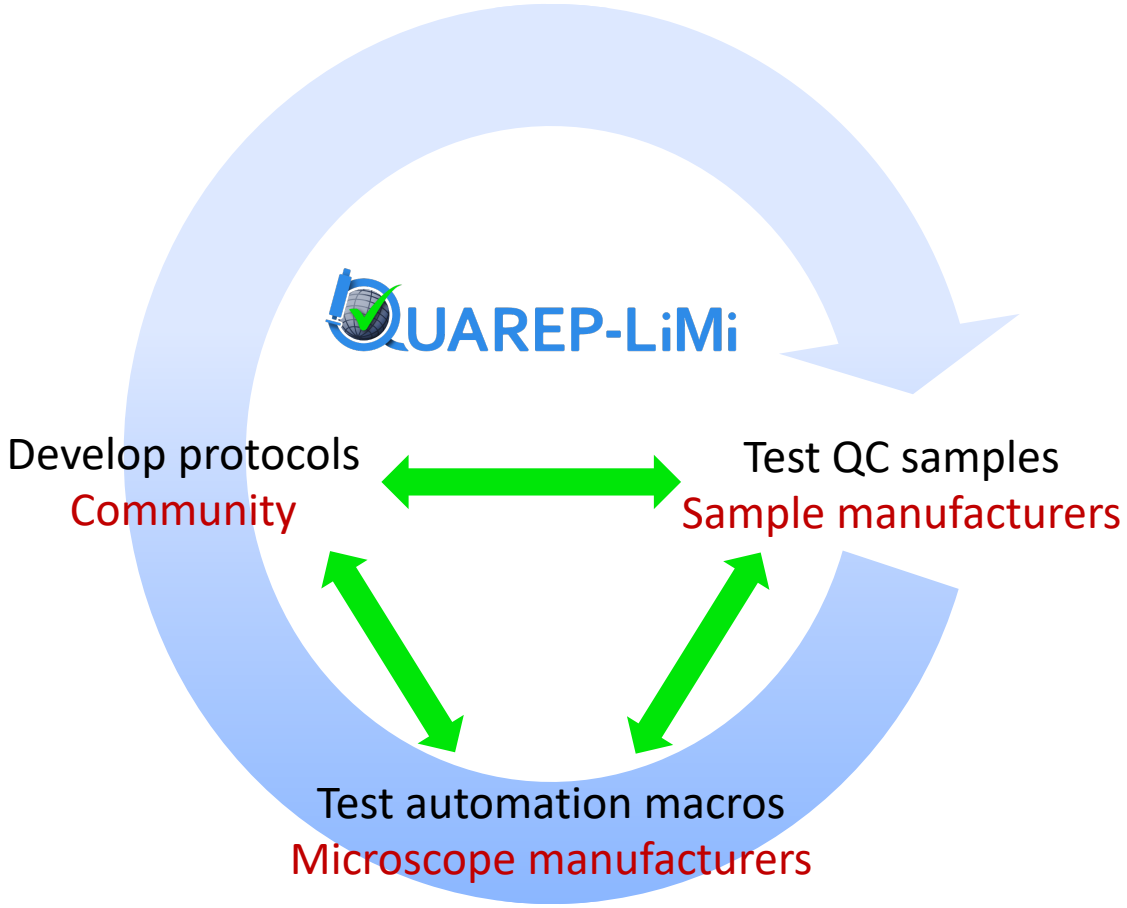
Check for updates

QUAREP-LiMi: a community endeavor to advance quality assessment and reproducibility in light microscopy

The community-driven initiative Quality Assessment and Reproducibility for Instruments & Images in Light Microscopy (QUAREP-LiMi) wants to improve reproducibility for light microscopy image data through quality control (QC) management of instruments and images. It aims for a common set of QC guidelines for hardware calibration and image acquisition, management and analysis.

Boehm U, **Nelson G**, et.al. QUAREP-LiMi: a community endeavor to advance quality assessment and reproducibility in light microscopy. Nat Methods. 2021 May 21. doi: 10.1038/s41592-021-01162-y. Epub ahead of print. PMID: 34021279.

QUAREP-LiMi



Working Groups
WG 1 ISO Illumination Power
WG 2 ISO Detector linearity and sensitivity
WG 3 ISO Uniformity of field - flatness
WG 4 ISO System chromatic aberration and Co-registration
WG 5 ISO Lateral and Axial Resolution
WG 6 ISO Stage and Focus – precision and other
WG 7 Metadata
WG 8 White paper
WG 9 Over all Planning + Funding
WG 10 Image Quality
WG11 Publication standards & methods

Further QUAREP-LiMi info/ publications

Join us!: https://quarep.org/contact

WG7:

Hammer et al., Towards community-driven metadata standards for light microscopy: tiered specifications extending the OME model. https://www.biorxiv.org/content/10.1101/2021.04.25.441198v3 (2021). Submitted to Nature Methods

Rigano et al., Micro-Meta App: an interactive software tool to facilitate the collection of microscopy metadata based on community-driven specifications. https://doi.org/10.1101/2021.05.31.446382 (2021). Submitted to Nature Methods

WG8:

Nelson, G, Boehm, U et al., QUAREP-LiMi: A community-driven initiative to establish guidelines for quality assessment and reproducibility for instruments and images in light microscopy. J. Microscopy 2021 Jul 2. doi: 10.1111/jmi.13041 https://onlinelibrary.wiley.com/doi/10.1111/jmi.13041

Boehm U, Nelson G, Brown CM, et.al.. QUAREP-LiMi: a community endeavor to advance quality assessment and reproducibility in light microscopy. Nat Methods. 2021 May 21. doi: 10.1038/s41592-021-01162-y. Epub ahead of print. PMID: 34021279.