OPTICALLY DETECTED MAGNETIC RESONANCE OF NANODIAMOND USING WIDEFIELD DETECTION

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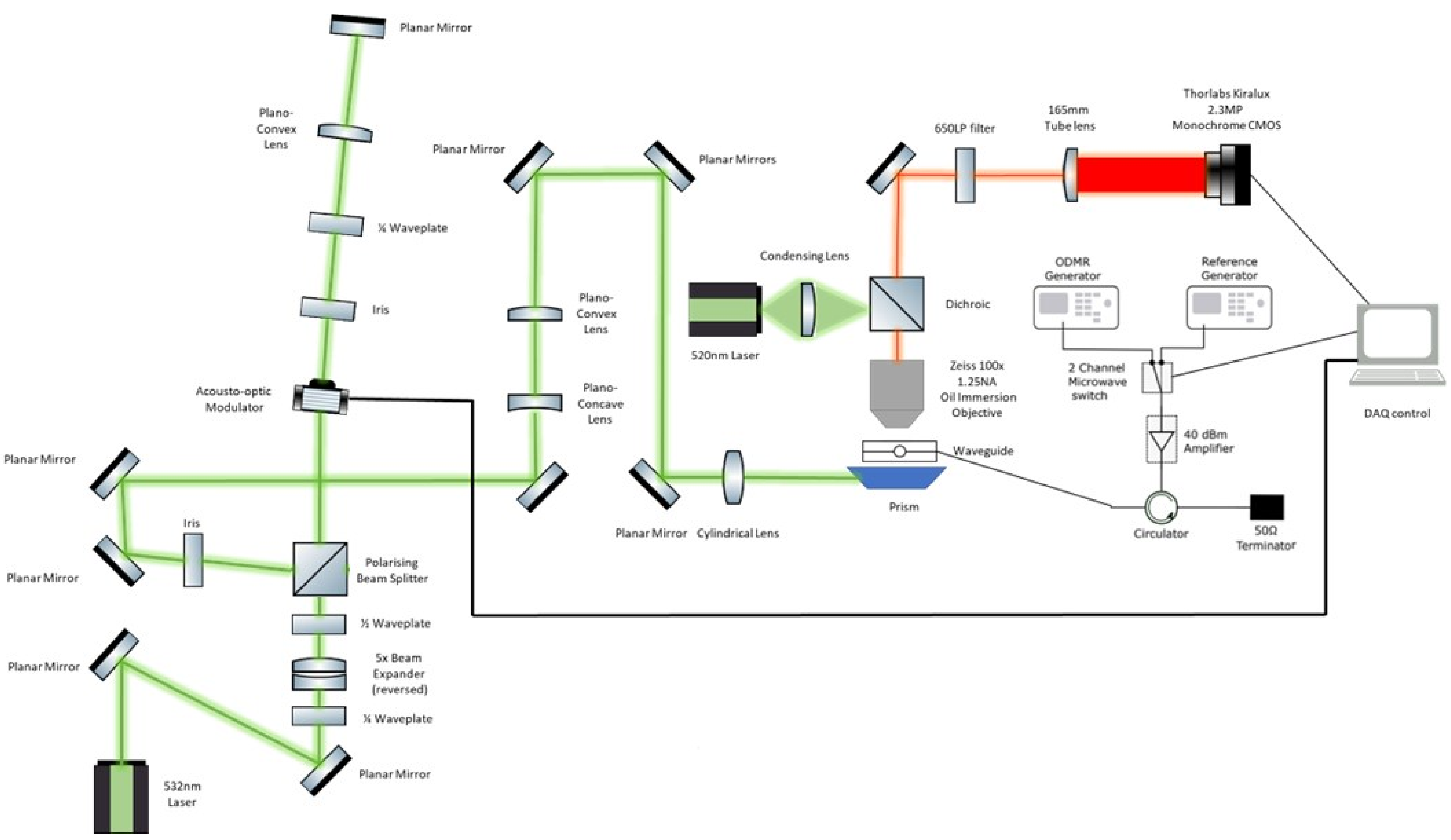
Optically Detected Magnetic Resonance (ODMR) imaging of fluorescent nanodiamond (FND) allows thermometry and magnetometry at a cellular level. It takes advantage of FND's biocompatibility and small size (between 5 and 100nm). Nitrogen-vacancy (NV) defects in the FND are optically excited at 532nm while applying a microwave field that is scanned around the resonant frequency of the triplet ground state of the NV (approximately 2.87GHz). There are two decay paths that offer differing emission intensities and that are coupled to the (microwave-controlled) spin state of the NV centre; by monitoring the change in fluorescence as a function of applied microwave frequency (the ODMR curve), the spin state of the NV centre can be inferred. Furthermore, this transition is both thermally and magnetically dependent; a magnetic shift sees a splitting of the detected response through the Zeeman effect and a thermal change sees a frequency shift that applies equally to all components of the curve.

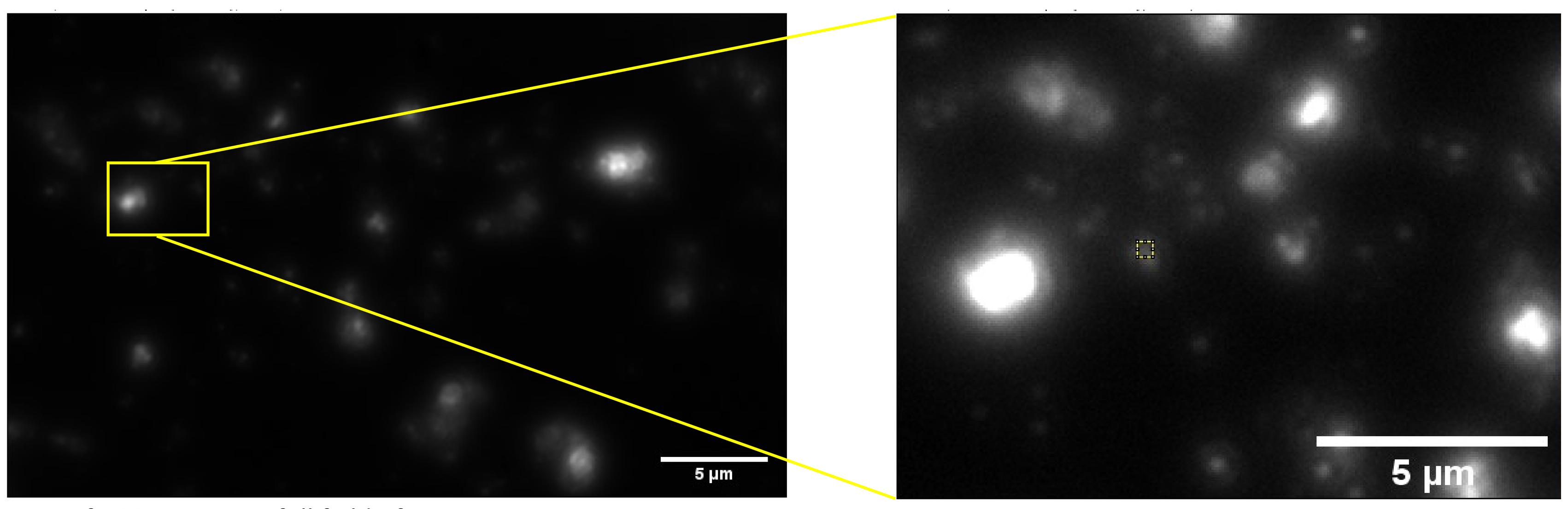
The Motivation

- FND's biocompatibility makes it a *brilliant* marker in biological samples, alongside it's photostability allowing long-term, high-powered excitation with no photobleaching effects.
- FND is typically bought from commercial suppliers and needs to be characterised to identify the batch's peak resonance frequency and other fluorescence properties. It would be ideal to do this on a system independent of the general research microscopes used for ODMR measurements in biologically relevant samples.
- When using FND as a bio-marker, widefield imaging allows us to get additional spatial information of the biological structures surrounding the FND. As a result, magnetometry and thermometry measurements can be linked to a biological or chemical function in the surrounds of the FND.
- Whilst epifluoresent microscopy is a generally suitable illumination method, Total Internal Reflection Fluorescence (TIRF) microscopy has the added benefit of only exciting the evanescent field of a few hundred nanometres above the coverslip. The entire sample isn't exposed to excessive illumination needlessly, which may be of benefit for photosensitive samples where the processes of interest occur close to the coverslip.

The Microscope



ODMR in Epifluoresence

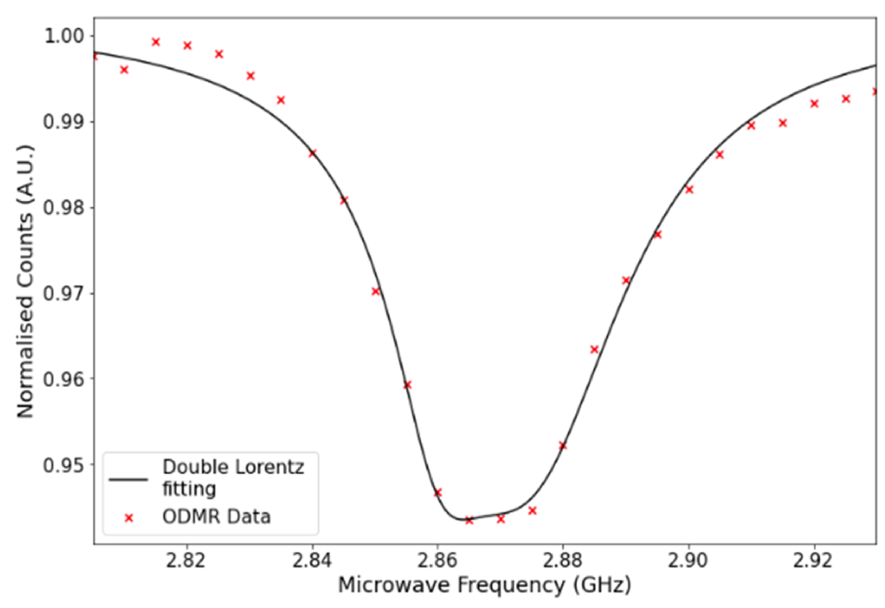


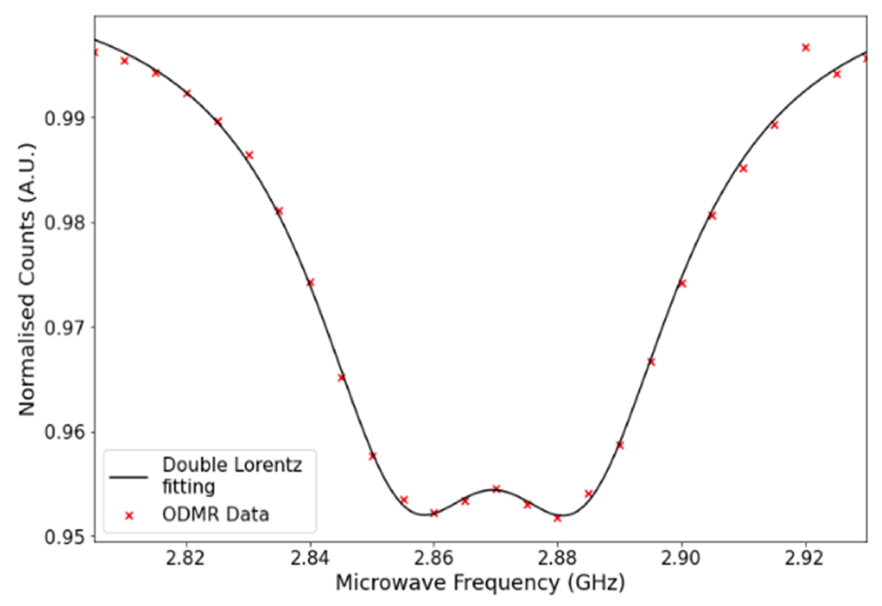
Top Left: 90nm FND in full field of view.

Top Right: Closeup of highlighted region. Due to the presence of large FND clusters, some regions are saturated: These are not analysed

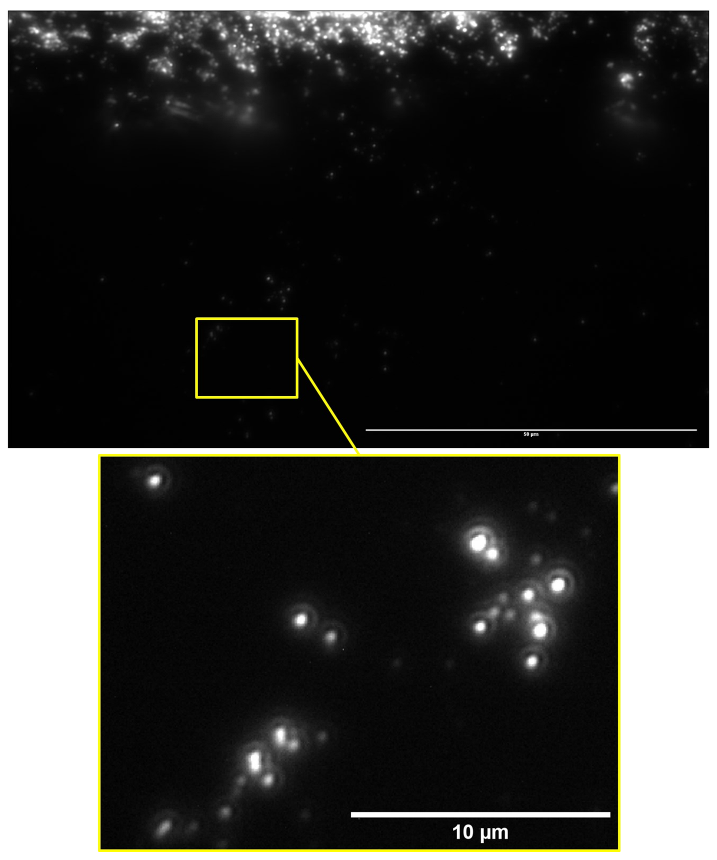
Bottom Left: Continuous wave frequency sweep 2.81-2.93GHz in steps of 5MHz.

Bottom Right: Continuous wave frequency sweep 2.81-2.93GHz in steps of 5MHz with magnetic field applied, Zeeman splitting evident.





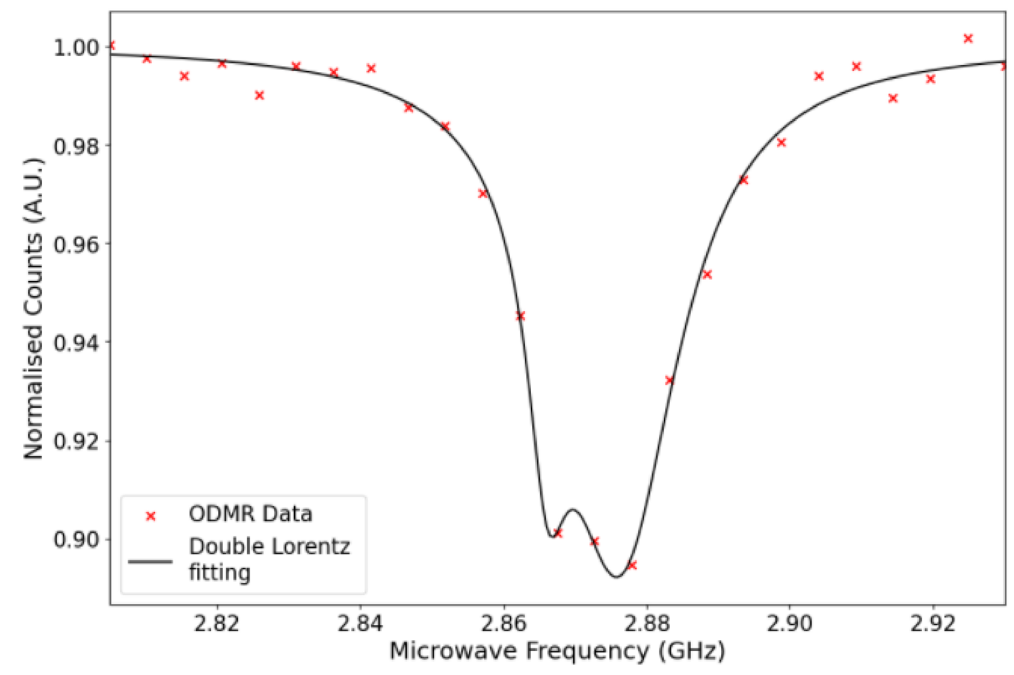
ODMR in TIRF



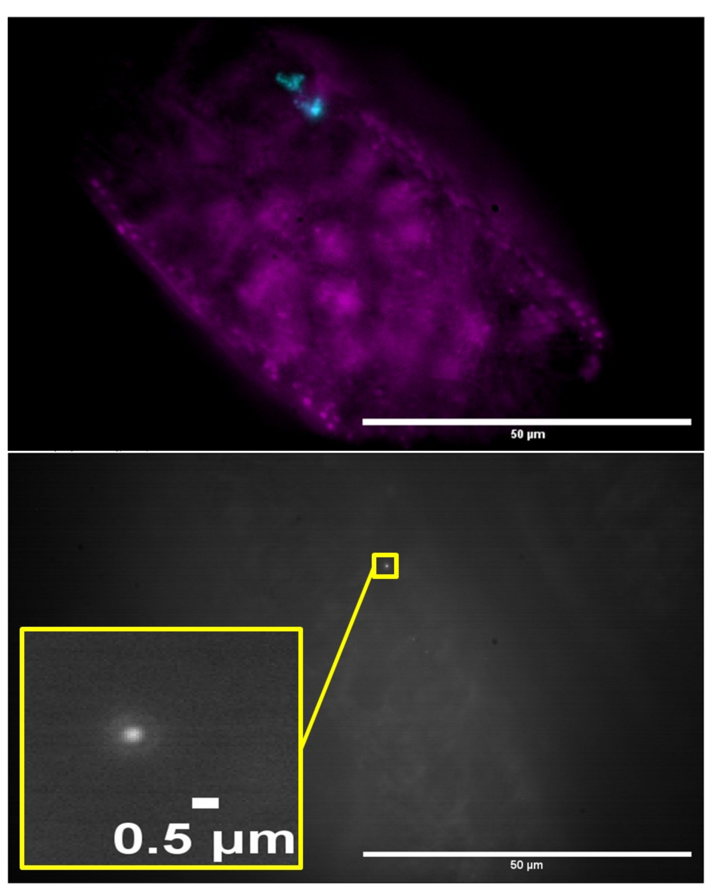
Top Left: 90nm FND in full field of view

Bottom Left: FND zoomed in to show Airy rings.

Right: Continuous wave frequency sweep 2.81-2.93GHz in steps of 5MHz.



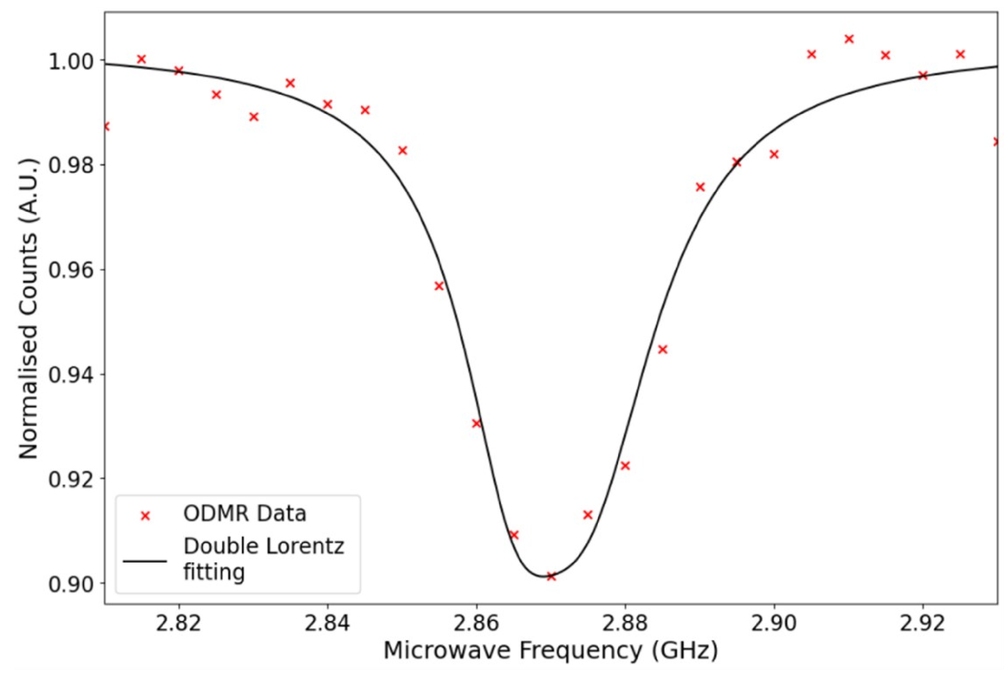
ODMR in *C. elegans*



Top Left: 90nm FND (cyan) in *C. elegans* labelled with mCherry (magenta) in epifluorescence microscopy.

Bottom Left: FND in same area, zoomed in to show Airy rings.

Right: Continuous wave frequency sweep 2.81-2.93GHz in steps of 5MHz.



Conclusions

- We have shown the proof of concept for a multifunctional widefield fluorescent microscope for ODMR measurements of FND's. This microscope has a resolution and field of view capable of biological imaging with a minimum contrast for ODMR of 6% , it also can detect magnetic field shifts.
- Further experimentation would involve relaxometery measurements and pulsed measurements, expanding the biological imaging into TIRF microscopy. Improving sample preparation to facilitate different biological samples would also be ideal.

References and Acknowledgements

[1] L. Rodin *et al.* 2014 *Rep. Prog. Phys.* **77** 056503

[2] Y. Nishimura *et al.* 2021 *Nature Sci. Rep.* **11** 4248

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EPSRC Engineering and Physical Sciences Research Council

NPL National Physical Laboratory

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