QUANTUM IMAGING

Super-resolution with quantum light

Quantum correlations from photon antibunching enhance the resolution of image scanning microscopy in biological imaging by twofold, four times beyond the diffraction limit.

Andrew Forbes and Valeria Rodriguez-Fajardo

Biological imaging traditionally uses bright classical light, where the wave nature of light places fundamental limits on the achievable resolution, which were set centuries ago by Rayleigh and Abbe. Despite the known advantages of quantum states of light for resolution-enhanced imaging, significant practical hurdles have prohibited its widespread implementation[1].

Reporting in *Nature Photonics*, Ron Tenne and colleagues now demonstrate a quantum version of the classical super-resolution method[2] of image scanning microscopy[3] (ISM), and apply it to image biological matter. Their approach combines the power of treating light as quantum particles (photons) with the structured illumination (wave) approach of ISM to achieve a fourfold resolution enhancement over the classical diffraction limit. Importantly, this is demonstrated in a conventional confocal microscopy set-up with only minor modifications, ushering in exciting prospects for practical quantum imaging of living matter.

Resolution in classical imaging has improved significantly over the past few decades, driven primarily by the demand of 'seeing smaller' in the biological sciences. Several super-resolution approaches have emerged that break the classical diffraction limit, doing so by violating at least one of the underlying assumptions made by Rayleigh and Abbe. Image scanning microscopy does so by using structured illumination rather than uniform illumination of the sample, offering a potential twofold resolution enhancement after some routine image processing. Quantum light too holds promise for resolution-enhanced imaging, including reaching sub-shot-noise levels[4] and probing with sparse photons[5], but has only seen limited success[6] in imaging biological matter because of the fragility of quantum states and the noisy environment of biological matter.

Now, Tenne and colleagues overcome these limitations by exploiting the antibunching property of single-photon emitters[7], showing that photon correlations in a biological sample can serve as the contrast of a super-resolved image[2]. The authors coin their approach quantum ISM (Q-ISM), a quantum-enhanced version of ISM for super-resolution quantum imaging (Fig. 1). They first excite the embedded quantum dots, at ~10 nm scale, in their samples, which are labelled microtubules in fixed cells, and then collect the emitted light with a bundled array of 14 fibres — a conventional (classical) ISM approach that is now commercially available and gives a twofold resolution enhancement over a conventional confocal microscopy set-up. But rather than only collecting the spatially resolved light through each fibre, they also consider the temporal resolution by observing coincidences of arriving photons at pairs of fibres, each fitted with small pixelated single-photon avalanche detectors (SPADs) with nanosecond temporal resolution. And therein lies the problem.

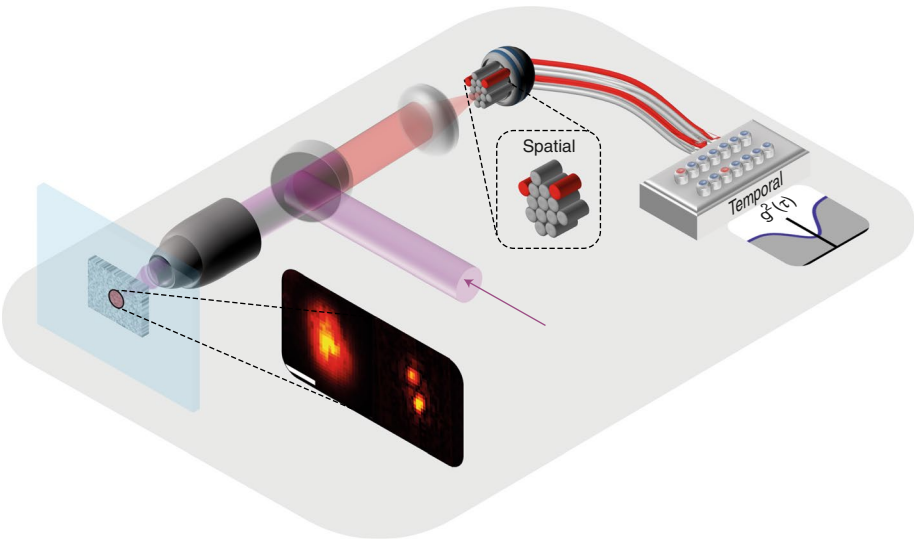

Fig. 1 | The Q-ISM approach to imaging. In Q-ISM, a conventional confocal microscopy set-up is modified by collecting light with a fibre bundle array and measuring the light collected (ISM), as well as the coincidences of arriving photons at pairs of fibres (an example pair is shown in red), thus providing spatial and temporal control. The purple arrow is the incoming light. The measured $g^2(0)$, the second-order intensity correlation function, from the photon coincidences can be used to find the missing photon pairs by subtracting it from that expected from Poisson statistics of the emitters. As a result of antibunching of the photons, the ISM point spread function is squared for a fourfold increase in resolution in Q-ISM, after some routine data processing. The inset shows images of single emitter quantum dots with a conventional scan (left) together with the processed Q-ISM image (right). Scale bar, 500 nm. The resolution enhancement is as a result of violating two of the Abbe–Rayleigh diffraction limit assumptions: using structured illumination while detecting quantum particles. Inset reproduced from ref. [2], Springer Nature Ltd.

It is well known in quantum ghost imaging[8] that the 'coincidences' hold tremendous information, far more in fact than the 'singles'. Likewise, in addition to the information in the singles of each fibre (the classical ISM data), the coincidences between pairs of fibres also contain valuable information. Unlocking this additional information yields the additional twofold enhancement in resolution, adding up to a fourfold overall enhancement[2].

Intriguingly, the key lies in looking for light that isn't there, what the authors call the missing photon pairs[2]. The quantum nature of light implies that each quantum dot emits only one photon, following sub-Poisson statistics. By detecting the photon coincidences between pairs of fibres, one finds missing photons, less than would be expected from a Poisson-like source. Using this information, the sample can be imaged and resolved even when the emitters are present in high density, almost completely overlapping and emitting identical photons, by virtue of the single-photon antibunching effect. This is because no emitter can ever produce two photons, even if it is excited by a source with two photons. Therefore, any coincidences must come from another emitter, regardless of proximity. The result is a nonlinear response at the detector, violating one of the diffraction limit assumptions and therefore allowing for a resolution enhancement. Using the ISM approach and the information in the missing coincidence signals (quantum) results in a fourfold enhancement in Q-ISM[2]. The advantage holds for the axial direction too, with the quantum signal falling off sharply with distance, providing an enhanced axial resolution. This makes Q-ISM suitable for z-sectioning of biological samples, which are often much thicker than the axial diffraction limit. In other words, the approach allows quantum-enhanced 'seeing' in three dimensions.

A key feature of Q-ISM is that it requires only minor modifications to an otherwise conventional microscopy set-up, replacing a pinhole with a small pixelated detector with good temporal resolution, alleviating the complexity synonymous with harnessing quantum states. Further, the low illumination levels necessary to achieve single-photon emission come with a most welcome advantage — it prevents fluorophores from bleaching as well as reduces the risk of photodamage to the biological matter, an issue faced by other super-resolution techniques, particularly stimulated emission depletion (STED) microscopy. Crucially, the Q-ISM contrast depends on the absolute number of missing photon pairs. It increases with the number of emitters, so that Q-ISM is more tolerant to the density of markers in the sample, meaning a higher density implies a better classical signal-to-noise ratio (SNR). This makes for a compelling case: good SNR (at low resolution) from the classical component of the imaging simultaneously with high resolution (but low SNR) in the quantum component.

There is yet room for improvement. Q-ISM requires long data acquisition times to overcome the low quantum SNR, is currently not wide field, and requires single-photon emitters making it incompatible with label-free approaches, such as stimulated Raman spectroscopy and photothermal spectroscopy. In future, it may be possible to merge ISM and Q-ISM data to form a classical–quantum hybrid image, since both are measured simultaneously and therefore describe the same scene, while recent advances in SPAD arrays[9], now with lower noise, faster response and with sizeable arrays of 512 × 512 pixels[10], could open the possibility of faster, wide-field Q-ISM.

The possibility of incorporating quantum measurements and well-known classical approaches opens up new imaging options for better insights into biological processes. Tenne and co-workers present a promising example of just such a situation for ISM, and one cannot help but wonder what other techniques could benefit from a similar approach. For instance, would it be possible to replace the detector used in STED microscopy in an analogous manner? The challenge is not to be underestimated, as it would be necessary to first develop single-photon sources that can be selectively deactivated.

Historically, quantum imaging systems have failed to achieve the performance of their classical counterparts, hindering their applications. Tenne and colleagues' advance makes clear that quantum imaging could add new information without sacrificing the classical information. They show hybrid imaging solutions for the best of both worlds. This suggests that practical quantum-enhanced imaging systems are now within sight, with the merger of Q-ISM and ISM opening a clear path to integration in a commercial device. ❐

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Integrated solution for quantum technologies

Integrated photonics could allow for the generation, manipulation and detection of quantum light on-chip, opening the path to a scalable, reliable platform for real-world deployment of quantum applications.

Rachel Won

About 500 attendees gathered in Nanjing, China from 9–11 November 2018 to participate in the Nature Conference on Nanophotonics and Integrated Photonics (NIP2018). It was organized by Nanjing University, *Nature Electronics*, *Nature Materials*, *Nature Nanotechnology* and *Nature Photonics*, and was the first Nature Conference in the field of optics and photonics. It featured 4 plenary talks and 35 invited talks in the areas of plasmonics, metamaterials, topological photonics, integrated photonics and quantum optics.

Of all the topics discussed, research into combining integrated photonics with quantum optics attracted much attention due to the surge in interest in