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Adaptive optical quantitative phase imaging based on annular illumination Fourier ptychographic microscopy

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Abstract
Quantitative phase imaging (QPI) has emerged as a valuable tool for biomedical research thanks to its unique capabilities for quantifying optical thickness variation of living cells and tissues. Among many QPI methods, Fourier ptychographic microscopy (FPM) allows long-term label-free observation and quantitative analysis of large cell populations without compromising spatial and temporal resolution. However, high spatio-temporal resolution imaging over a long-time scale (from hours to days) remains a critical challenge: optically inhomogeneous structure of biological specimens as well as mechanical perturbations and thermal fluctuations of the microscope body all result in time-varying aberration and focus drifts, significantly degrading the imaging performance for long-term study. Moreover, the aberrations are sample- and environment-dependent, and cannot be compensated by a fixed optical design, thus necessitating rapid dynamic correction in the imaging process. Here, we report an adaptive optical QPI method based on annular illumination FPM. In this method, the annular matched illumination configuration (i.e., the illumination numerical aperture (NA) strictly equals to the objective NA), which is the key for recovering low-frequency phase information, is further utilized for the accurate imaging aberration characterization. By using only 6 low-resolution images captured with 6 different illumination angles matching the NA of a 10x, 0.4 NA objective, we recover high-resolution quantitative phase images (synthetic NA of 0.8) and characterize the aberrations in real time, restoring the optimum resolution of the system adaptively. Applying our method to live-cell imaging, we achieve diffraction-limited performance (full-pitch resolution of 655 nm at a wavelength of 525 nm) across a wide filed of view (1.77 mm²) over an extended period of time.

Keywords: Quantitative phase imaging; Fourier ptychographic microscopy; Adaptive optics; Long-term imaging

Introduction
Quantitative phase imaging (QPI) is a powerful optical tool for visualizing and quantifying optical thickness variation of unlabeled transparent biological samples without the need for specific exogenous contrast agents [1, 2, 3]. Various QPI techniques have been developed in the past decades and achieved great success in biomedical applications. These techniques include digital holography (DH) [4, 5, 6], phase shifting interferometry [7, 8], lateral shearing interferometry [9, 10], transport-of-intensity equation (TIE) [11, 12, 13], differential phase contrast (DPC) [14, 15, 16],
Fourier ptychographic microscopy (FPM) [17, 18, 19, 20], among others. Many of these techniques are performed using a modified microscope system. The imaging throughput follows the law of Lagrange invariant [21], leading to the unavoidable trade-off between imaging resolution and field-of-view (FOV). Among them, FPM is a promising solution to achieve high-throughput QPI by synthesizing a wide-field, high-resolution complex image from multiple low-resolution images captured under angle-varied illumination. It has been demonstrated for long-term label-free observation and quantitative analysis of large cell populations without compromising the spatial and temporal resolution [19, 22, 23, 24, 25]. Free from the adverse effects of staining reagents on cell viability and signaling, high-throughput QPI can find important applications in drug discovery, personalized genomics, cancer diagnosis, and drug development [26, 27, 28, 29].

Nevertheless, high spatio-temporal resolution imaging over a long-time scales (hours to days) and across a wide FOV remains a critical challenge [30, 31, 32], due to the spatially non-uniformity and temporally varying system aberrations. The contributing factors of this challenge include optically inhomogeneous structure of biological specimens, mechanical instability of the microscope, and thermal fluctuations of the environment [33]. Their combined impact can significantly degrade the achieved resolution and quality of the imaging platform. Moreover, the aberrations are sample- and environment-dependent, and thus they cannot be compensated using a fixed optical design [34]. Therefore, an adaptive method for the dynamic correction of wavefront distortions over a large FOV is highly desired in QPI systems.

Adaptive optics (AO) was originally developed for astronomical telescopes [35, 36]. This technology can detect the aberrations introduced by the atmosphere and correct them using a deformable mirror. Nowadays, the AO-based technology has found extensive applications in optical imaging [37, 38], particularly in confocal and multiphoton microscopy, as it facilitates three-dimensional volumetric imaging of thick biospecimens [39, 40]. One major drawback of the AO technology is the need for additional hardware, such as Shack-Hartman wavefront sensor, spatial light modulator, and deformable mirror. In contrast, with the help of inherent data redundancy, FPM enables direct aberration recovery and correction post measurement. As a result, it can reconstruct a high-quality image without resorting to AO hardware [41, 42, 43], offering a solution as “computational” AO [44, 45]. However, the data redundancy requirement (generally dozens of images) in conventional FPM significantly limits its imaging speed, resulting in low temporal throughput. Moreover, the adaptive wavefront correction in FPM is generally designed for imaging absorptive samples. Their applicability to QPI of transparent phase objects remains to be explored.

In this article, we propose an adaptive optical QPI (AO-QPI) method to tackle the temporally- and spatially-varying aberrations for live-cell imaging. Based on the annular illumination FPM (AIFPM) [22], the aberration and phase can be efficiently decoupled from the captured intensity images through nonlinear optimization. Here, we demonstrate the adaptive aberration correction without the need for traditional AO hardware. Compared with the conventional aberration correction scheme in FPM, AO-QPI only needs 6 images taken under matched illuminations to
recover the pupil aberration. To demonstrate the validity and practicability of AO-QPI, we performed long-term live-cell imaging using our prototype setup, achieving diffraction-limited performance across a FOV of 1.77 mm², a time during of 51 hours and a frame rate of 16.66 Hz (can reach the camera speed of 100 Hz as the sliding window scheme is adopted). To the best of our knowledge, it is the first time to perform dynamic aberration correction on QPI to secure the high-quality imaging performance over time, especially at such a high spatiotemporal bandwidth.

Materials and methods

Optical setup and principle

The reported AO-QPI system was built using a standard bright-field microscope [IX73, Olympus, Japan, see Fig. 1(a)] with a programmable LED array as the illumination source. During the image acquisition process, the LED elements were sequentially turned on to illuminate the specimen from different angles. We then acquired the corresponding low-resolution images using a 10×, 0.4 NA objective lens (UPLSAPO10×, Olympus). The specimen phase as well as the wavefront aberrations were coupled in the captured intensity images, which need to be recovered through nonlinear optimization algorithm. Figure 1(b) shows the workflow of the reconstruction process, which takes the cell specimen as an example for demonstration. The initial guess of the specimen is obtained by a coherent version of DPC deconvolution [15]. The captured images are then successively adopted to impose the intensity constraints for jointly updating the specimen and the pupil function. Here, the pupil function is characterized by the Zernike polynomials for an efficient and accurate recovery. When the convergence condition is met, the specimen phase and the aberration are recovered from the raw images. As is shown in Fig. 1(d), through the reconstruction process mentioned above, the intracellular details are resolved with a high resolution after ∼50 iterations.

It seemed that the reconstruction process follows a similar framework of FPM with aberration correction. However, compared with traditional FPM methods which usually require dozens of frames as raw data, the AO-QPI has two significant improvements for enhancing the imaging efficiency, including the optimization of illumination strategy and aberration correction algorithm. By integrating the two innovations, AO-QPI is able to realize dynamic aberration correction with one order of magnitude fewer measurements.

Illumination mode optimization

In the traditional FPM imaging process, a dense sampling strategy in the Fourier domain is adopted to meet the requirement of the spectrum overlapping rate [46]. While in the method of AO-QPI, based on the optical transfer function theory [22], we only need several raw images taken with the illumination NA matching the objective NA. Such an illumination condition has been verified to be significant for phase retrieval in asymmetric-illumination-based QPI methods [16, 47, 48]. For a practical imaging system with non-negligible aberrations, the object spectrum and system aberrations are coupled as two inseparable components in the captured images and the matched illumination condition is equally crucial for accurate aberration recovery, which has been validated in Supplementary Note 3. To address
the speed-accuracy tradeoff for imaging, the minimum data redundancy required for a successful aberration characterization is further discussed in Supplementary Note 4. We finally choose to cyclically turn on 6 NA-matched LED elements (see Fig. 1(c)) to acquire the raw dataset for dynamic phase imaging. Benefiting from the successful reconstruction with such small amount of raw images, the adaptive correction is sustained over the high-speed imaging process to defy the temporal-varying aberrations in real time. Moreover, we also apply a sliding window along the whole captured image sequence, thus the effective phase imaging frame rate is further raised to the shutter speed of the camera.

Aberration recovery algorithm

The common reconstruction algorithms of FPM [41, 49, 50] are derived from several versions of ptychographic iterative engine (PIE) [51, 52, 53], which reconstruct the complex amplitude of specimen by alternating iterations in the spatial and frequency domains. The updating process for the specimen spectrum is given as follows,

\[ O = O + \alpha \left( \frac{|P|^2}{P_{\text{max}}^2} \right) \left[ \frac{P^*}{P} \right]^2 [\Phi_u - \Phi_e] \]

(1)
where, $O$ is the spectrum of the specimen, $P$ is the pupil function of the optical system, $\Phi_u$ and $\Phi_e$ are the spectrum with and without the intensity constraint, and $\alpha$ is the updating step size. Moreover, for the redundancy of the captured raw images, the pupil function can be reconstructed in a similar way as shown in Eq. 2, where the pupil function and the spectrum are reversed. The updating formula is first proposed in the method of EPRY [41], which is a modified version of ePIE method [54] applied in FPM.

\[
P = P + \beta \frac{|O|^2}{|O|_\text{max}^2} \frac{O^*}{|O|^2} [\Phi_u - \Phi_e]
\]  

(2)

Here, we report a revised pupil function updating formula, as shown in Eq. 3. The term of normalized spectrum modulus $|O|^2/|O|_{\text{max}}^2$ is omitted in the new formula. For the extremely high energy of the DC term in the bright-field spectrum, the term $|O|^2/|O|_{\text{max}}^2$ is close to zero and will make the pupil function $P$ hard to be effectively updated. The similar normalized term was adopted for the update of probe function in PIE [51] and ePIE [54] to defy the detector noise. However, it is not applicable to FPM because the object function $O$ in the update formula is transformed from the spatial domain to the frequency domain. The comparison of the reconstruction results using Eq. 2 and Eq. 3 are provided in Supplementary Note 1.

\[
P = P + \beta \frac{O^*}{|O|^2} [\Phi_u - \Phi_e]
\]  

(3)

In practical implementations, Eq. (1) and Eq. (3) are applied to alternatively update the object function and the pupil function until the convergence condition is satisfied. However, the joint estimation of both object and pupil requires more raw dataset for obtaining the correct solution. In order to achieve aberration correction while ensuring imaging efficiency, the prior knowledge of the common aberrations is introduced to the reconstruction process. Here, we adopted the Zernike polynomials $Z_m^n$ [55, 56, 43] as a suitable basis to express the pupil aberration. Therefore, the degrees of freedom in solution space is reduced from a two-dimensional matrix to a small number of coefficients associated with the dominant Zernike modes, which is beneficial to obtaining a correct reconstruction with limited raw images. More details and improvements of this implementation are further discussed in Supplementary Note 4, which is adopted as a way to reduce the requirement for data redundancy.

**Samples for testing**

We tested our method on a standard microlens array, a phase resolution target and HeLa cell cultures in Petri dishes. The micro lens array (18-00036, SUSS, Microoptics, refractive index (RI) of 1.45) was placed on a cover glass for imaging. The curvature radius is 9670 µm and the pitch is 130 µm for this target. Its theoretical maximum height is 0.22 µm, corresponding to a maximum phase of $\sim$1 rad. The adopted quantitative phase microscopy target (QPT$^TM$, Benchmark Technologies
Corporation, USA) provides an optical path difference for QPI by increasing the thickness of the bars on a glass plate. The bars are made of the acrylate polymer (The refractive index is 1.52) on Corning Eagle XG Glass including groups 6 - 10 with the feature height of 150 nm. The cells were cultured in a 37°C incubator with 5% CO2 for observation to further demonstrate the performance of the proposed method on biospecimens.

Computation platform used for AO-QPI
The AO-QPI reconstruction algorithm was performed based on MATLAB software (MATLAB R2018b) with a laptop equipped with a 2.60 GHz central processing unit (Intel Core i7-10750H) and 16 GB of random-access memory. As we mainly focused on the proof-of-concept, the image reconstruction speed was not optimized. The time required for the computation processes (including 50 iterations to solve the phase and aberration simultaneously) of the raw intensity images (2048 × 2048 × 6) is ~ 20 minutes. In the computation processes, we divided each full-FOV raw image (2048 × 2048 pixels) into 10 × 10 sub-regions (256 × 256 pixels each), with a 60-pixel overlap on each side of neighboring sub-regions. After completing the reconstruction of each sub-region, a full-FOV result is generated by using an alpha-blending stitching method. The consuming time for the reconstruction could be further reduced by implementing GPU acceleration.

Results and discussion
Optimal illumination scheme for AO-QPI
In this section, we presented our simulation study and experimental results to validate the illumination scheme of AO-QPI for the accurate and efficient aberration characterization. The simulation parameters were chosen to realistically model the experimental platform. Figure 2(a) shows the input specimen phase and imaging aberration for simulation, where a micro lens is adopted as the simulated specimen and the aberration is represented by a certain combination of the Zernike polynomials. In Supplementary Note 2, we have verified that the annular illumination mode could replace the traditional FPM illumination mode for an effective ptychographic reconstruction with much fewer raw images, which is followed by simulations in this section. Figure 2(b) further demonstrates the necessity of the matched illumination condition for the accurate characterization of both phase and aberration. We first performed the reconstruction using 12 raw images with the matched illumination, and the obtained results are almost identical to the input, as shown in the first row of Fig. 2(b). For a fair comparison, we also adopted the same number of raw images for reconstruction, but with the illumination NA half of the objective NA in the second row of Fig. 2(b). In this case, the recovered aberration has a similar distribution but weaker amplitude compared to the input. It indicates that there are still uncorrected aberrations that could degrade the image recovery. The corresponding recovered phase shows a circular outline which is a typical result obtained by high-pass filtering. This is due to the fact that the low-frequency information is lost in the imaging process without the matched illumination condition. The results in Fig. 2(b) verified the necessity of the matched illumination configuration for a correct reconstruction (the detailed analysis are provided in
Supplementary Note 3). To explore the minimum dataset required for a successful aberration characterization, we provide the convergence curves as well as the reconstructed pupil aberrations using different number of raw images in Figs. 2(c) and 2(d). Here, we adopted the algorithm of Zernike mode constraint to improve the performance with limited number of raw images. For comparison, the comparable results with the method of EPRY are provided in Supplementary Note 4. As shown in Fig. 2(d), when only three raw images are adopted, the aberration error (RMSE) curve initially descends in the first several iterations but rebounds rapidly, and finally converges to a stable value with some fluctuations. The obtained result in Fig. 2(c) completely deviates from the input which can be attributed to the insufficiency of data redundancy. The increase in data redundancy could steer the algorithm to better convergence but the results are still unsatisfactory. As the number of raw images increases to more than six, the RMSE curve achieves a solid decline and finally converges to near zero, which indicates that we have successfully characterized the aberration.

As a further validation of the forementioned conclusions, we performed an experiment on the micro lens array, which was deliberately placed at an out-of-focus position. The LED array was adjusted to an appropriate height so that the six LED elements in the array can exactly provide the annular matched illuminations for imaging (the coordinates of these LED elements in the array are (0, ±5), (3, ±4), (-3, ±4), in Fig. 2(e2)). We first sequentially lit 121 LED elements distributed in an array to acquire the raw dataset. In Fig. 2(e1), all raw images are utilized for a typical FPM reconstruction and the imaging aberration as well as the phase of a micro lens are successfully recovered. The obtained aberration is mainly composed of a spherical aberration caused by defocusing, and the remaining part represent the error of the imaging system. Then we excluded most of the raw images and only adopted six of them to satisfy the matched illumination condition for reconstruction and the results are provided in Fig. 2(e2). As we have demonstrated in simulations, the aberration and phase obtained in this way are almost identical to the standard result. Figure 2(e3) provides the reconstructed results using 37 raw images with the illumination angles smaller than the objective NA. As with the results in the simulation, the aberration and phase fail to converge to the correct solution in this case. It indicates that more raw dataset will not help the correct solution without the matched illumination. Based on the simulation and experiment results, it can be concluded that the matched illumination configuration provides an efficient way for the accurate reconstruction of both sample phase and pupil aberration, and six raw images could provide sufficient data redundancy for the correct solution under this condition.

AO-QPI on a quantitative phase microscopy target
A quantitative USAF phase resolution target was imaged to quantify the AO-QPI’s capacity to restore the diffraction-limited resolution in the presence of imaging aberration. To better demonstrate the aberration correction performance, the QPT was deliberately placed at an out-of-focus position to magnify the imaging aberrations. We first perform a standard FPM method without aberration correction to perform the quantitative phase reconstruction using totally 121 raw images. However,
Figure 2 Reconstructed phase and aberration under different illumination modes. (a) Simulated specimen phase (above) and aberration (below). (b) Reconstructed results with and without matched illumination, including illumination modes of the LED array (the red circle is NA of the objective lens), recovered aberrations, recovered phase and corresponding phase error. (c) Reconstructed results with 3, 6, 9 images. (d) Convergence curves of the reconstruction with different number of images. (e) Reconstructed phase of microlens in the full FOV. (e1) Experiment result of ROI using full 121 images. (e2) Experiment result of ROI using 6 images under the matched illumination. (e3) Experiment result of ROI using 37 images without the matched illumination.
due to the uncorrected imaging aberrations, significant defocus artifacts appear on the recovered phase (Fig. 3(a)), which severely degrade the obtained imaging resolution. Then, the method of EPRY was performed using the same set of raw images, which is a common algorithm for aberration correction in FPM. With the characterization of the imaging aberrations, a desired phase map is successfully obtained, as is shown in Fig. 3(b). The minimum center-to-center distance of the line profile across the bars demonstrate the conservatively estimated resolution of 690 nm, which is consistent with theoretical resolution of the imaging system (655 nm, 0.8 NA). The proposed AO-QPI was equally tested for the same recovery of both the imaging aberration and specimen phase as shown in Fig. 3(c). By integrating the annular matched illumination scheme and the aberration correction algorithm based on Zernike mode constraint, AO-QPI could provide the comparable result with only six raw images. Moreover, through the comparison of the reconstructed aberrations in Fig. 3(b) and 3(c), we can see that a real aberration could be appropriately characterized by the Zernike modes we adopted in the reconstruction algorithm. Through the experimental results above, we can see that the method of AO-QPI provides an efficient way to remove the aberrations and achieve the diffraction-limited performance of the imaging system.

AO-QPI on HeLa cell cultures for a full-field imaging
In this section, the AO-QPI method was applied for imaging HeLa cell cultures across a full FOV of 1.77 mm². Although a flat-field microscope objective was adopted in the experiment, the optical imaging tends to exhibit aberrations that vary across the field. To defy the spatially-varying aberrations, we divided the full
FOV into 10 × 10 sub-regions and performed the AO-QPI reconstruction on each region independently. In Figs. 4(b)-4(g), we provide the recovered cell phase of six selected regions, as well as the recovered aberrations. In the provided phase maps, the subcellular structures are revealed with high-quality imaging performance, which could be adopted as a proof that the aberrations within each local FOV have been effectively characterized and corrected. To further demonstrate the necessity of our method, we also compared phase maps reconstructed with and without the aberration correction. Figures 4(f1)-4(g1) are the zoomed-in views of cells in the red and blue box of Figs. 4(f) and 4(g). With the successful characterization of the aberration, the punctate ribosome structures inside the cells can be clearly distinguished with the resolution approaching the theoretical value of the imaging system (see Fig. 4(f3) and 4(g3)). In Fig. 4(f2) and 4(g2), the comparable results are reconstructed without the aberration correction except for an auto-refocusing algorithm [57] to remove the effect of defocus. It can be seen that the subcellular structures with the size close to the diffraction limit have been completely smoothed out due to the uncorrected aberrations. These results demonstrate the capacity of our approach to sustain the high-quality imaging performance over a large field, suggesting that it provides a powerful tool for high-throughput biological analysis. More dynamic results to further verify this point can be seen in Visualization 1.

AO-QPI on HeLa cell cultures over an extended period of time
The AO-QPI method was further performed for continuous observation of HeLa cell cultures for an extended period of time, which is a challenging biomedical application for microscopy. The adverse factors for long-term imaging can be attributed
to thermal fluctuations of the environment [58], mechanical instability of the system [59], and evaporation from the cell media [60]. All these factors will lead to the temporal-varying aberrations across the whole imaging process and significantly degrade the imaging quality. To solve these problems, we apply the AO-QPI method to provide the adaptive aberration correction and continuously acquire the high-quality phase maps across 51 hours. The obtained time-lapse movie is provided in Visualization 2. Figure 5(a) shows the full-FOV phase map stitched together from the two frames at hour 0 and hour 51. After the reproduction over dozens of hours, the cell populations went from being sparsely distributed to almost covering the full FOV. From Fig. 5(a1) to 5(a5) are the five frames of the zoomed-in cell phase maps extracted from the movie at a 12-hour interval. The corresponding recovered aberrations vary significantly across the whole imaging process and the main reason can be attributed to focus drift (see the line chart beside). In Figs. 5(b1)-5(b4), we demonstrate the performance of AO-QPI to trace the variation of subcellular features across a shorter time frame. In the provided four frames of enlarged images, we could clearly observe the varying details inside a single cell at different time points. In Figs. 5(c1)-5(c3), to further verify the necessity of our method, we compare the cell phase maps reconstructed with the same six raw images but corrected by the aberrations recovered at different time points. It can be seen from the obtained phase maps that a fixed correction could not sustain the imaging quality for even few hours, and the adaptive correction scheme is hence indispensable for long-term imaging. Benefiting from the long-term stable imaging capacity of AO-QPI, we further study the cell morphology during cell divisions covering three generations. The cell images at the key timings are given in Figs. 5(d1)-5(d6). The mother cell in Fig. 5(d1) underwent three mitoses across \(\sim 27\) hours and eventually divided into four individual daughter cells, as shown in Fig. 5(d6). The provided experiment results reveal the capacity of the AO-QPI approach for correcting temporally varied aberrations and securing the imaging performance for long-term longitudinal study.

Through the experimental results demonstrated in Fig. 5, we can see that the proposed AO-QPI provides a computational solution for adaptively correcting the disturbances in long-term imaging process, without involving additional hardware. Compared with the existing methods [61, 62, 59, 63] for tracking focus drift in real time, our approach is flexible and allows digital refocusing after the data has been acquired. In addition, the commonly used algorithm of gradient detection for autofocusing is problematic for cell culture dishes as the intensity image has the lowest contrast when the sample is placed at the in-focus position. The reported AO-QPI scheme can address this challenge and offer a turnkey solution for long-term in vitro imaging in a robust and stable manner.

**Conclusions**

In this article, we first apply an FPM-based computational AO method in QPI to secure the high-quality performance for long-term imaging across a wide field. The proposed AO-QPI approach is based on the annular matched illumination configuration, which has been verified to be essential for the recovery of low-frequency phase information [22, 48]. Here, through the simulations and experiments, we further reveal that such an illumination configuration is equally indispensable for the
Figure 5 Long-term live-cell phase imaging across 51 hours [see Visualization 2]. (a) The Full-FOV phase maps of the first and last frame in the movie. (a1)-(a5) The zoomed-in phase maps in the central region of (a) as well as the line chart indicating the out-of-focus distance variation over the 51 hours. (b1)-(b4) Dynamic high-resolution demonstration of cellular architecture across 4 hours. (c1)-(c3) Comparison of results with and without adaptive aberration correction as well as the profiles of the cell phase. (d1)-(d6) The 3-generation cell division process from 8h 6min to 35h 26min.
accurate characterization of the imaging aberrations. This can be attributed to
the characteristic of the aberration which mainly consists of low-frequency com-
ponents. To improve the imaging efficiency, we also analyze the minimum data
redundancy required for a successful recovery of both phase and aberration. We
find that only six raw images could provide sufficient raw dataset for the ptych-
ographic reconstruction, which could satisfy the requirement for real-time imaging.
For a further demonstration of AO-QPI’s performance on biomedical samples, we
apply the method on HeLa cell cultures for a long-term and wide-field imaging. The
diffraction-limited imaging performance is secured across a large FOV of 1.77 \( mm^2 \),
over a long period of 51 hours at 16.66 \( Hz \) frame rate. Our demonstration indicates
that the AO-QPI approach offers a powerful tool for joint spatiotemporal adaptive
aberration correction.

The FPM-based aberration correction approach has been widely applied in many
challenging fields to improve the imaging performance [42, 64, 65, 66, 67]. However,
a limitation is that these works are mainly aimed for stained samples. In this article,
we first apply the similar framework for quantitative phase imaging. It should be
noted that due to the transfer response difference of sample’s phase and absorption
components in FPM imaging process [22, 48], the aberration recovery for pure phase
objects is more difficult to be achieved based on a normal FPM illumination scheme.
The adopted annular matched illumination configuration could provide a reliable
and efficient approach for aberration correction on the transparent biospecimen.
Moreover, as an extension to FPM, the technology of 3D FPM [68, 69, 70, 71]
is developed to further reveal the 3D phase information for thick samples. The
existence of aberrations could also affect the imaging performance of the 3D phase
imaging. Therefore, applying AO-QPI to achieve the high-quality depth-resolved
imaging for thick phase object is another important direction that requires further
investigation.

Abbreviations
QPI: Quantitative phase imaging; FPM: Fourier ptychographic microscopy; NA: Numerical aperture; DH: Digital
holography; TIE: Transport-of-intensity equation; DPC: Differential phase contrast; AO: Adaptive optics; AI:
Annular illumination; FOV: Field of view; LED: Light-emitting diode; PIE: Ptychographic iterative engine; ePIE:
Extended ptychographic iterative engine; EPRY: Embedded pupil function recovery; QPT: Quantitative phase
microscopy target; 3D: Three-Dimensional.

Declarations
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Authors’ contributions
C.Z. and J.S. provided the original idea. Y.S. and J.S. designed and performed the experiments. Y.S. and Y.F.
analyzed the data. Y.S., N.Z. and R.Y. prepared the figures and supplementary movies. Y.S. and J.S. wrote the
manuscript. C.Z., Q.C. and G.Z. supervised the overall projects. All the authors read and approved the final
manuscript.

Availability of data and materials
The datasets used and analysed during the current study are available from the corresponding author on reasonable
request.
Ethics approval and consent to participate
There is no ethics issue for this paper.

Competing interests
The authors declare that they have no competing interests.

Consent for publication
All authors agreed to publish this paper.

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