High-Speed Fourier Ptychographic Microscopy System for Live Microorganisms Imaging

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Abstract: We present a high-speed Fourier ptychographic microscopy system with a custom LED module that is 535 times brighter than an off-the-shelf board, achieving 150 fps live microorganisms imaging and reducing motion blur. © 2025 The Author(s)

1. System Designing for High-Speed Fourier Ptychographic Microscopy

Acquiring high-speed dynamic scenes at high resolution and large field-of-view (FOV) is in high demand in biological and medical research. Fourier ptychographic microscopy (FPM) achieves large FOV and high spatial resolution by illuminating the sample at various angles [1]. However, FPM struggles to capture dynamic scenes due to the long total acquisition time by both a large number of patterns and long exposure time for each pattern, resulting in low temporal resolution. To address these limitations, previous works have improved the acquisition speed of FPM from both algorithmic and hardware perspectives–such as illumination multiplexing [2] and hardware enhancement [3].

In this work, we propose a high-performance FPM system featuring a custom LED module with 535 times higher effective brightness compared to a standard off-the-shelf LED board, enabling high-speed live microorganisms imaging. The system synchronizes the LED module with a high-speed camera and employs snapshot illumination multiplexing for acquisition and a space-time algorithm for reconstruction [4]. Please see Fig. 1 for an illustration of the overall system design.

Our key insight into system design is that achieving high-speed acquisition requires maximizing the number of photons received by the sensor per unit time, which we refer to as effective brightness. This allows shorter exposure time and results in higher frame rate. However, the inefficiency of traditional LED control algorithms is one of bottlenecks limiting effective brightness—a typical off-the-shelf LED panel connects LEDs in a matrix manner with shared pins for rows and columns and utilizes a dynamic refreshing algorithm, where only a subset of LEDs are illuminated at a time. The controlling system iteratively illuminates all subsets to display one pattern, which is called refreshing. This approach reduces the effective brightness of each LED and causes flickering artifacts when the sensor's exposure time is shorter than the refreshing period, limiting its applicability in high-speed imaging.

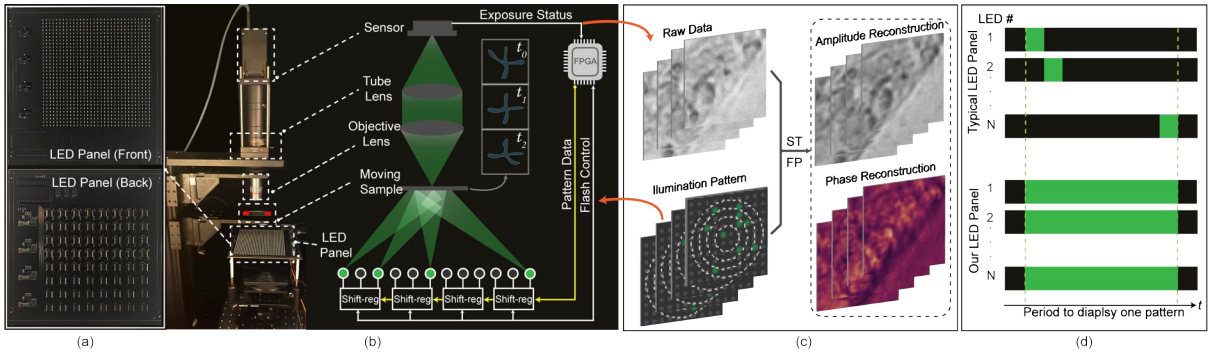

Fig. 1. (a) Front and back views of our custom LED module. (b)–(c) Overview of the proposed FPM system, with (b) the hardware setup and (c) the snapshot acquisition and space-time reconstruction algorithm [4]. (d) Comparison of two LED controlling algorithms. Green (black) indicates the on (off) status of each LED. The relative effective brightness of each LED is 1/N in a typical off-the-shelf LED board and 1 in our approach.

To overcome these limitations, we adopt an array of shift-register LED drivers, where each pin is dedicated to controlling a single LED. This configuration allows the system to simultaneously illuminate all LEDs based on

preloaded data, eliminating the requirements of dynamic refreshing, which maximizes the effective brightness of each LED. Furthermore, shift-register chips usually feature a double buffer design, enabling new data to be loaded while displaying a pattern. As a result, the fastest achievable frame rate is determined solely by the data transfer speed, significantly enhancing system frame rate. Please refer to Fig. 1 for a comparison.

We further select the brightest green LEDs available on the market within a narrow wavelength bandwidth. This maximizes the signal-to-noise ratio (SNR) and minimizes chromatic aberration. We also choose a high-frame-rate camera and adopt illumination multiplexing algorithm to achieve high-speed acquisition.

2. Implementation & Experiment Results

We implemented a prototype setup as illustrated in Fig. 1, consisting of a custom LED board with the previously described control algorithm, a high-frame-rate camera, and optical components. Here the space-time optimization for FPM [4] is used for illumination multiplexing and reconstruction.

The LED board is placed 80 mm from the sample plane, providing 88% spectrum overlap. Light transmitted through the sample is collected by a 10× objective (Mitutoyo M Plan Apo, 0.28 NA), relayed via a tube lens (Thorlabs TTL200-S8) to a LUCID Atlas10 camera (ATX081S-MC, 136.7 fps at 8.1 MP).

To implement our LED control algorithm, we utilize TLC5927 constant LED sink drivers as shift-register. A total of 64 drivers are daisy-chained to control 1,024 LEDs, forming a 32×32 matrix with a 4 mm pitch. The LED model is ASCKCG00-NW5X5020302 with dominant wavelength at 520 nm. Each LED has 1.8 cd luminous intensity, enabling short exposure acquisition, minimizing motion blur. We utilize a Cyclone 10 LP FPGA, operating at a 48 MHz system clock, to push patterns to the board and synchronize the LED board and camera. Illumination patterns are transferred from the PC to the FPGA via UART. The shortest illumination time in our implementation is 21 ns, and the maximum LED board frame rate is 31,250 fps.

We demonstrate the effectiveness of our system through a live microorganism experiment using brine shrimp. Specifically, we compare results obtained under a low-fps configuration (15 fps, 55 ms exposure) and a high-fps configuration (150 fps, 1 ms exposure), as shown in Fig. 2. The two cases are configured by setting the LED driver current to its minimum and maximum, resulting in different LED brightness and exposure times, while keeping all other parameters fixed. The low-fps condition is a typical setting in FPM systems with off-the-shelf LED boards. As shown, the high-fps capturing significantly reduces motion blur compared to the low-fps case, preserving fine spatial details of amplitude and phase. We also compare brightness between our LED module and an off-the-shelf board (Adafruit P4-2121-16S-HL1.0). Using a calibrated linear camera, we capture identical patterns at the same distance and normalize intensities by exposure time. Ours provides 535 times higher effective brightness.

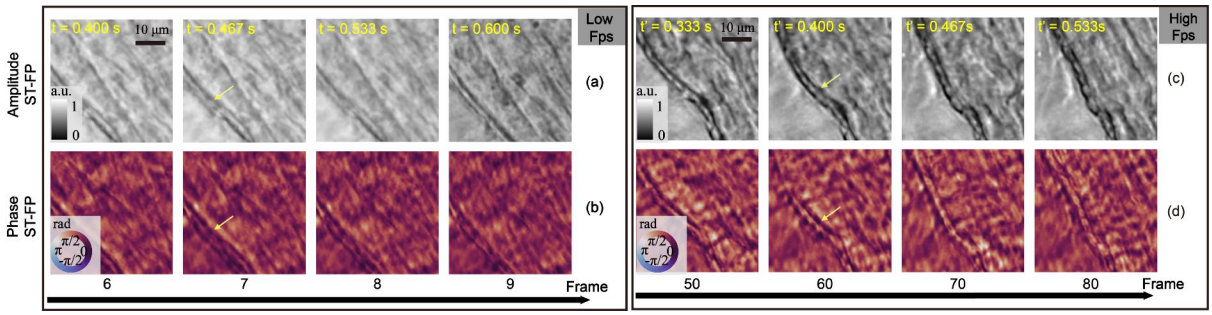

Fig. 2. Comparison of (a)-(b) low-fps and (c)-(d) high-fps live brine shrimp imaging. The region shown corresponds to the same internal structure under fast physiological movement, with its boundary indicated by yellow arrows. In the low-fps case, boundary and internal structures are blurred due to rapid motion. In contrast, the high-fps case results preserve sharpness and fine details.

References

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