Super-resolution Microsphere Digital Holographic Microscopy Imaging based on Phase-intensity Feature Fusion

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ABSTRACT

Microsphere digital holographic microscopy (MDHM) is an emerging label-free, non-invasive imaging technology that combines the optical properties of microspheres with digital holographic imaging, and has shown great potential in the biomedical field. MDHM can utilize the super-resolution characteristics of microspheres to simultaneously obtain the phase and intensity information of the sample, providing a more detailed perspective for cell morphology research and disease diagnosis. However, limited by the diffraction limit of the optical system and the sampling rate of the camera, the resolution of MDHM images often cannot meet the needs of fine observation of cell microstructures. To solve this problem, we develop a super-resolution reconstruction method based on phase-intensity feature fusion. It has different feature extractors for phase and intensity characteristics, and achieves feature fusion through the attention mechanism, which effectively improves the reconstruction effect. Experimental results show that our method achieves excellent performance in the super-resolution reconstruction tasks of normal red blood cells (nRBC) and thalassemia red blood cells (tRBC), laying a foundation for the further application of MDHM in the biomedical field.

Keywords: Feature fusion, digital holography, super-resolution

1. INTRODUCTION

Digital holography (DH) senses the amplitude and phase information of the object and processes the recording holograms using computational algorithms. Combined with deep learning, DH could achieve material characterization and classification, high-throughput detection, high-resolution imaging, and 3D tracking.[1–7] Microsphere digital holographic microscopy (MDHM) combines the optical properties of microspheres with the interference principle of digital holographic microscopy (DHM),[8,9] and has received widespread attention in the biomedical field in recent years.[10] The uniqueness of microspheres lies in their superlens effect: through micron-sized spherical particles, the focusing ability of the imaging system can be greatly improved, breaking through the diffraction limit of traditional optical systems and achieving super-resolution imaging.[11] Specifically, the microspheres focus the light passing through them to the subwavelength scale, so that the details that were originally indistinguishable can be clearly captured on the sensor.[12] In addition, MDHM can not only obtain the phase and intensity information of the sample in a label-free and non-invasive manner, but also more effectively amplify the microscopic details, revealing the subcellular level structures and subtle pathological changes that are difficult to observe with traditional DHM.[13] In the MDHM imaging process, the microspheres act as super-resolution lenses, and their powerful optical focusing characteristics greatly improve the resolution of the optical system. Through digital holographic reconstruction, the enhanced phase and intensity information of the microspheres can be extracted from the hologram, further revealing the internal structure and surface characteristics of the sample.[14,15] Therefore, MDHM is regarded as a powerful super-resolution imaging technique, providing a new approach for cell morphology analysis,[16] and it has potential for disease diagnosis.

Although MDHM has significant advantages, its resolution is still limited by system parameters, such as the size of the microspheres, the refractive index, and the sampling rate of the camera. In addition, in actual imaging, the diffraction limit of the optical system, the light loss in microsphere imaging, and noise interference will affect

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the image quality. Therefore, when studying microstructures, the imaging effect of MDHM may still be subject to some challenges, especially when detecting extremely small subcellular structures or subtle pathological changes. Traditional methods to improve imaging resolution include optimizing microsphere materials, using shorter wavelength light sources, or increasing the numerical aperture of the system, but these methods usually require expensive optical components and complex experimental conditions, making them difficult to be widely used.

To overcome these problems, super-resolution reconstruction methods based on deep learning have brought new possibilities for MDHM image enhancement. Unlike traditional DHM, MDHM imaging not only contains rich phase and intensity information, but also has super-resolution characteristics and optical effects introduced by microspheres, so a special network structure is required to fully explore these features. However, existing deep learning super-resolution methods mainly target natural images, ignoring the unique phase-intensity information of MDHM and the wrapping characteristics and nonlinear characteristics of phase images. In addition, the phase and intensity information in MDHM images are often closely related. How to make full use of the complementarity of the two to improve the reconstruction quality is an urgent problem to be solved.

This study proposes a super-resolution reconstruction method based on phase-intensity feature fusion, which aims to make full use of the advantages and complementarity of the phase and intensity information of MDHM to improve the resolution and reconstruction quality of the image. We specifically designed different feature extractors for phase and intensity characteristics to deeply capture the optical properties introduced by the microspheres; at the same time, we introduced an attention mechanism to adaptively integrate phase and intensity features to achieve high-quality fusion and joint reconstruction of phase and intensity information. To verify the effectiveness of this method, we conducted experiments on normal red blood cells (nRBC) and thalassemia red blood cells (tRBC) samples.[17] The results showed that this method has significant advantages in maintaining physical consistency and improving imaging quality.

2. METHOD

2.1 Data Processing

This study used a microsphere digital holographic image dataset[17] containing 760 normal red blood cells (nRBC) and 609 thalassemia red blood cells (tRBC). Each image has a size of 1024×1280 pixels and contains 3 color channels. After digital reconstruction, these microsphere holograms can obtain phase and intensity information respectively, thereby further exploring the cell microstructure.

2.1.1 Hologram Reconstruction

To extract phase and intensity information from the original microsphere hologram, we used a digital reconstruction method based on Fourier transform. This method uses the optical magnification effect of microspheres and can perform digital processing while preserving the imaging characteristics of microspheres. First, the color microsphere hologram is converted into a grayscale image to eliminate the interference between color channels. Then, the grayscale image is subjected to a two-dimensional Fourier transform to convert the spatial domain information into the frequency domain. In the frequency domain, we apply a transfer function to simulate the propagation of light waves in space. This step is based on diffraction theory and takes into account physical parameters such as wavelength and propagation distance. Next, the processed frequency domain signal is converted back to the spatial domain by inverse Fourier transform to obtain a reconstructed wavefront in complex form. Finally, the phase and intensity information are extracted from this complex wavefront respectively. The phase information is obtained by calculating the argument of the complex number, while the intensity information is obtained by calculating the modulus of the complex number.

2.1.2 Low-resolution Downsampling

To simulate the low-resolution effect in the actual imaging process, we processed the reconstructed high-resolution phase and intensity images. The high-resolution image was scaled down to reduce the resolution using the bicubic interpolation method. This step simulates the diffraction limit of the optical system and the limited sampling rate of the camera. Then, Gaussian noise was added to the downsampled image to simulate various noise sources in the imaging system, such as photon noise, readout noise, etc. Finally, the processed low-resolution image was interpolated back to the original size, keeping the consistency of the input and label sizes. This step simulates

the upsampling operation commonly used in practical applications. In order to facilitate network training and ensure numerical stability, we normalized the phase and intensity images and mapped their value range to the range of [0,1]. This operation not only helps to accelerate network convergence, but also reduces interference between features of different scales.

2.2 Network Design

2.2.1 Overall Architecture

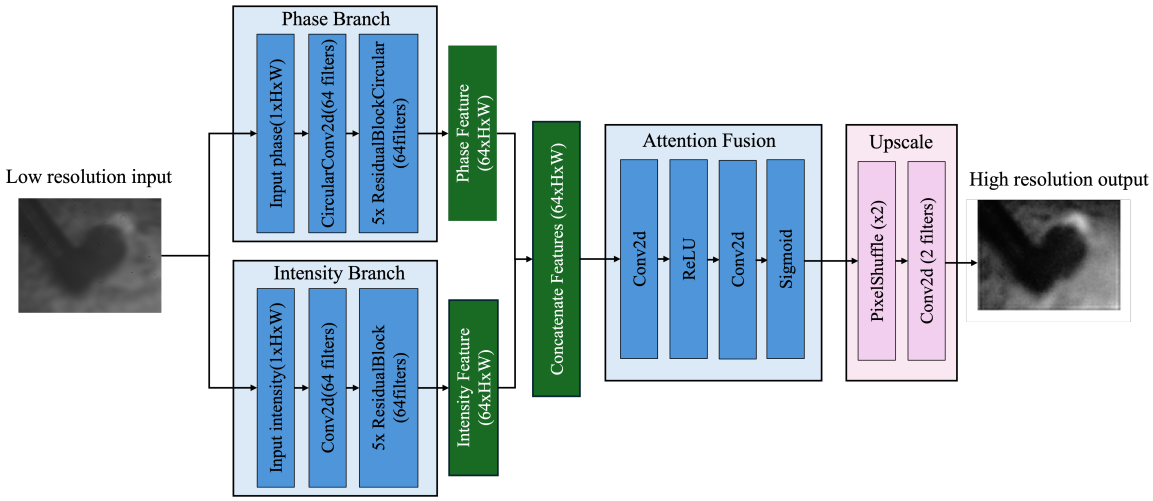

Figure 1. Schematic of the network structure.

Based on the characteristics of microsphere digital holographic microscopy, we proposed a network architecture consisting of four main components: phase branch, intensity branch, feature fusion layer and reconstruction layer. The phase branch designs a feature extractor using circular convolution and residual blocks to address the periodicity of microsphere phase images, the wrapped phase characteristics, and the optical effects introduced by microspheres. This design fully considers the characteristics of MDHM images and aims to effectively extract and fuse phase and intensity information. The phase branch designs a feature extractor using circular convolution and residual blocks based on the periodicity and wrapped phase characteristics of the phase image. Circular convolution can effectively handle phase jumps and maintain phase continuity. The introduction of residual blocks helps the network learn deeper feature representations while alleviating the gradient vanishing problem. The intensity branch uses conventional convolutional layers and residual blocks to focus on extracting edge and texture features of intensity images. This design takes into account the similarity between intensity images and natural images and can effectively capture local structural information. The feature fusion layer uses the attention mechanism to fuse the phase and intensity features. The attention mechanism can adaptively highlight important features and suppress irrelevant information, thereby achieving more effective feature integration. The reconstruction layer converts the fused features into high-resolution phase and intensity images through upsampling and convolution layers. This design ensures the end-to-end learnability of the reconstruction process and can adaptively adjust the reconstruction strategy according to task requirements. The schematic of the network structure is shown in Fig. 1.

2.2.2 Loss function

We use the weighted L1 loss function to calculate the reconstruction errors of phase and intensity respectively, focusing on the phase-intensity characteristics of the microsphere digital holographic image, such that

$$\mathcal{L}_N = \lambda_{\text{intensity}} \cdot \left\| \hat{I} - I \right\|_1 + \lambda_{\text{phase}} \cdot \left\| \hat{\phi} - \phi \right\|_1 . \tag{1}$$

Here, I and ϕ are the intensity and phase images reconstructed by the network, I and ϕ are the high-resolution true values, and $\lambda_{\text{intensity}}$ and λ_{phase} are weight parameters. In this study, we set both weight parameters to 1 to give equal importance to phase and intensity reconstruction.

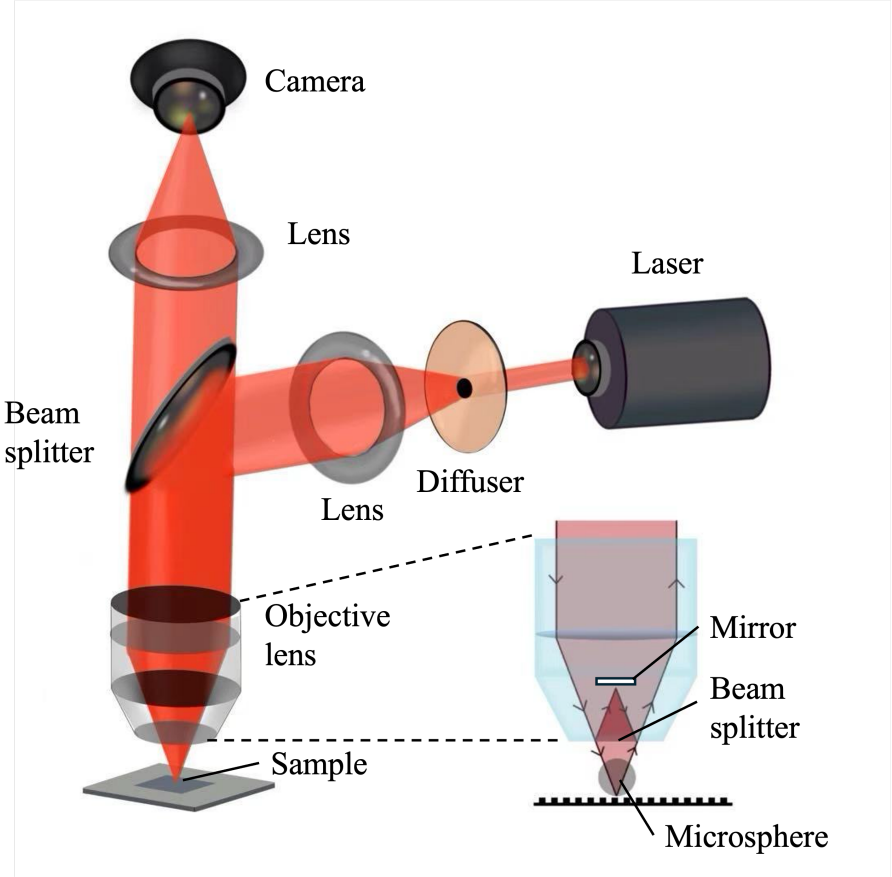

Figure 2. Schematic diagram of the MDHM system. A zoomed-in diagram shows the details of the optical path at the sample plane.

2.2.3 Training Strategy

This paper uses the Adam optimizer with an initial learning rate of 10^{-4}. The Adam optimizer combines the advantages of the momentum method and the adaptive learning rate method and can effectively handle sparse gradients and non-stationary targets. The batch size is set to 4. Considering the high-resolution characteristics of the image and the limitations of computing resources, a smaller batch size can improve memory utilization while ensuring training stability. The number of training rounds is set to 50 rounds. Through experimental observation, we found that 50 rounds of training can make the model fully converge while avoiding overfitting. We dynamically adjust the learning rate according to the performance of the validation set. Specifically, we adopt a learning rate decay strategy with a decay factor of 0.1. When the performance of the validation set does not improve for 5 consecutive epochs, the learning rate is reduced to 0.1 times the original. This strategy can fine-tune the model parameters in the later stage of training and improve the generalization ability of the model. To improve the generalization ability of the model, we use a variety of data augmentation techniques, including random rotation ($0°, 90°, 180°, 270°$), horizontal and vertical flipping, and slight brightness and contrast adjustments. These enhancement methods can simulate the changes in the actual imaging process and enhance the adaptability of the model to different imaging conditions.

2.3 Experimental Setup

We show the schematic diagram of an MDHM system in Fig. 2. A microsphere is accurately placed at the top of light beam.

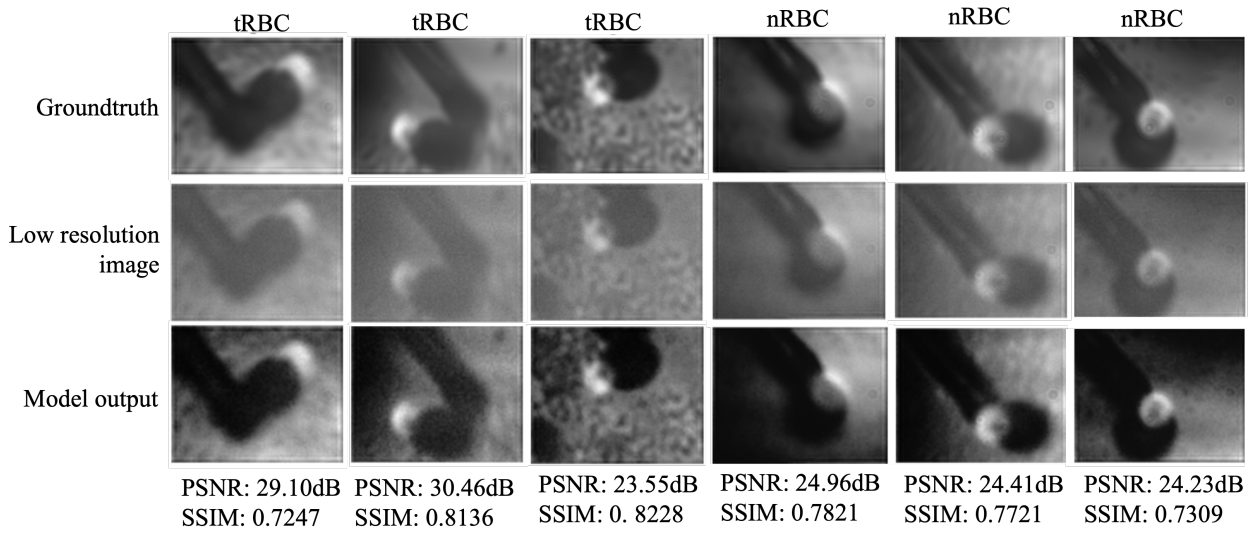

Figure 3. Experimental results for the super-resolution microsphere digital holographic reconstruction with phase-intensity feature fusion.

2.4 Dataset Preparation

We divided the dataset into training, validation, and test sets in a ratio of 8:1:1. Specifically, the training set contains 1095 images (608 nRBCs and 487 tRBCs), and the validation and test sets each contain 137 images (76 nRBCs and 61 tRBCs). This division ensures that the ratio of nRBCs and tRBCs in each subset is consistent, which helps the model learn the characteristics of different types of cells.

3. EXPERIMENTS

3.1 Quantitative Evaluation

To comprehensively evaluate the reconstruction quality, we adopted two widely used image quality evaluation indicators: peak signal-to-noise ratio (PSNR) and structural similarity (SSIM). These two indicators are used to evaluate the reconstruction quality of phase and intensity images, respectively. PSNR mainly reflects the pixel-level difference between the reconstructed image and the original image. The higher the value, the better the reconstruction quality. It is particularly effective for evaluating the overall reconstruction accuracy. SSIM pays more attention to the preservation of structural information and can better reflect the human eye's perception of image quality. The closer the SSIM value is to 1, the more similar the reconstructed image is to the original image in structure. The combined use of these two indicators can comprehensively evaluate the quality of the reconstruction results, taking into account both pixel-level accuracy and the preservation of structural information, providing a reliable basis for method evaluation.

On the test set, as shown in Fig. 3, our method achieved the highest PSNR and SSIM indicators in both phase and intensity image reconstruction tasks. Phase image reconstruction: The average PSNR reaches 25.42 dB and the SSIM is 0.7944. Intensity image reconstruction: The average PSNR reaches 21.91 dB and the SSIM is 0.6520.

3.2 Qualitative Evaluation

Through visual comparison, the images reconstructed by our method excel in the following aspects. The outline of the cell membrane is clearer and the details of the internal structure of the cell are richer. Noise suppression, the background area is smoother and the noise interference is significantly reduced. Contrast enhancement, the contrast between the cell and the background is improved, which is conducive to subsequent analysis and recognition. Structural consistency, the structural information of the phase and intensity images is highly consistent, reflecting the good preservation of physical information during the reconstruction process. These observations

verify the effectiveness of our network design and prove the complementarity of phase and intensity information, and the important role of feature fusion.

4. DISCUSSIONS AND CONCLUSIONS

Phase images are periodic and discontinuous, which requires a specific convolution method to effectively extract features. The introduction of circular convolution is to solve this problem. In contrast, the texture and edge information of intensity images are closer to natural images, so they are suitable for using traditional convolution structures. This targeted design can better capture their respective characteristics. Although phase and intensity information come from the same sample, they reflect different aspects of the cell. Phase information mainly reflects the internal structure and optical thickness of the cell, while intensity information reflects more surface features and scattering properties. Fusion of these two complementary information can provide a more comprehensive representation of cell structure, thereby improving the accuracy and robustness of reconstruction.

Our network design fully considers the physical properties of microsphere digital holographic images, especially the periodicity of phase images in microsphere imaging,[18] the wrapped phase characteristics,[19] and the optical magnification effect brought by microspheres. Based on the characteristics of microspheres, the reconstructed phase and intensity images have higher resolution and quality. This design based on physical principles not only improves the reconstruction quality, but also enhances the interpretability of the model, making the reconstruction results more reliable and credible. The branch structure of the network and the introduction of the attention mechanism make the entire architecture have good scalability. This modular design allows us to add new feature extraction branches or fusion strategies as needed, making it possible to fuse more types of information (such as fluorescence information) in the future. Our method achieves end-to-end learning from low-resolution input to high-resolution output. This design avoids the complex pre-processing and post-processing steps in traditional methods, simplifies the entire super-resolution reconstruction process, and improves the practicality of the method.

The main contributions of this research include: First, a novel network architecture is proposed that can effectively process the phase and intensity information of MDHM images. Second, circular convolution is introduced to process phase images to solve the problems of phase jump and periodicity. Third, a feature fusion strategy based on the attention mechanism is designed to achieve adaptive integration of phase and intensity information. Fourth, this method surpasses existing methods in multiple evaluation indicators and sets a new benchmark for MDHM image super-resolution reconstruction.

There are still some limitations and directions for future improvement in this study. For example, the current method is mainly optimized for red blood cell samples, and further adjustments may be required for other types of cell or tissue samples. In addition, although our method significantly improves image resolution, there is still room for improvement in performance under extremely low signal-to-noise ratio conditions. Future research directions include introducing more physical models and prior knowledge[15,20] to further improve the accuracy and interpretability of reconstruction. Explore the incorporation of time series information into the reconstruction process to achieve high-resolution imaging of dynamic cellular processes. Combine information from other sensor modalities (such as terahertz metasurface, microwave) to achieve multimodal super-resolution reconstruction.[21–25] Develop dedicated models for specific diseases or cell types to improve performance in specific application scenarios. In general, this study provides an effective new method for super-resolution reconstruction of MDHM images, paving the way for further development in the field of computational imaging.

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