Optics Communications 402 (2017) 104–108

Contents lists available at ScienceDirect

Optics Communications

journal homepage: www.elsevier.com/locate/optcom

Enhanced resolution in lensless in-line holographic microscope by data interpolation and iterative reconstruction

Shaodong Feng, Mingjun Wang, Jigang Wu *

Biophotonics Laboratory, University of Michigan—Shanghai Jiao Tong University Joint Institute, Shanghai Jiao Tong University, Shanghai 200240, China

ARTICLE INFO

Keywords:
Compact lensless imaging
Digital holography
Phase retrieval

ABSTRACT

In a compact lensless in-line holographic microscope, the imaging resolution is generally limited by the sensor pixel size because of the short sample-to-sensor distance. To overcome this problem, we propose to use data interpolation based on iteration with only two intensity measurements to enhance the resolution in holographic reconstruction. We did numerical simulations using the U.S. air force target as the sample and showed that data interpolation in the acquired in-line hologram can be used to enhance the reconstruction resolution. The imaging resolution and contrast can be further improved by combining data interpolation with iterative holographic reconstruction using only two hologram measurements acquired by slightly changing the sample-to-sensor distance while recording the in-line holograms. The two in-line hologram intensity measurements were used as a priori constraint in the iteration process according to the Gerchberg–Saxton algorithm for phase retrieval. The iterative reconstruction results showed that the iteration between the sample plane and the sensor planes can refine the interpolated data and thus further improve the resolution as well as the imaging contrast. Besides numerical simulation, we also experimentally demonstrated the enhancement of imaging resolution and contrast by imaging the U.S. air force target and a microscope slide of filamentous algae.

1. Introduction

Compact microscopic imaging techniques are very attractive for many potential applications in healthcare and biology because of their low-cost and portable system design [1–6]. Among the reported techniques, digital lensless in-line holographic microscope (LIHM) [7–10] has the advantages of achieving wide field-of-view imaging with flexible sample-to-sensor distance and the capability of acquiring multilayer sample images simultaneously. Conventional in-line holographic reconstruction (will be called "direct reconstruction", as opposed to "iterative reconstruction" below) based on scalar optical field propagation generally has two issues that affect the imaging resolution and image quality, i.e., twin-image background caused by the loss of phase information and the reduced resolution in the sampling process of the imaging sensor.

Many methods have been proposed to eliminate or reduce the twin-image background [11–24]. Iterative phase retrieval techniques [11,12] are among the most popular solutions, where the phase information of the optical field is retrieved iteratively according to multiple intensity measurements [14–16,19,22] or one intensity measurement [17,21,23,24]. Once both intensity and phase information of the light field are known, the holographic reconstruction will be free of twin-image and thus have better SNR and image contrast.

On the other hand, the imaging resolution in LIHM is mainly determined by two factors: the detection numerical aperture (NA) and the sensor pixel size (Δ). Physically, the ultimate resolution limit will be the Abbe diffraction limit, i.e., ~0.61 λ/NA in Rayleigh criterion, where λ is the wavelength of illumination light. Thus the resolution can be improved by enlarging the NA of the optical system when the wavelength is fixed [25–29]. For compact in-line holography setup, the sample-to-sensor distance is very small (~5 mm in our experiment) and the sensor size is relatively large ($5.7 * 4.3$ mm in our experiment), so the NA will be large enough to achieve submicron resolution according to the Abbe diffraction limit. However, the pixel size (Δ) of the commercially available imaging sensor is generally larger than 1.2 μm because of fabrication and sensitivity requirements. So the resolution in LIHM will be limited by the pixel size of the sensor because there is almost no magnification for short sample-to-sensor distance. To overcome the sampling issue caused by the relatively large sensor pixel size, researchers proposed many effective techniques, for example, the optofluidic microscope (OFM) [30–34], where the sampling distance

* Corresponding author.
E-mail address: jigang.wu@sjtu.edu.cn (J. Wu).

http://dx.doi.org/10.1016/j.optcom.2017.05.059
Received 15 March 2017; Received in revised form 19 May 2017; Accepted 21 May 2017
0030-4018/

and imaging resolution is decreased by tilting an aperture array with aperture size smaller than the sensor pixel size, and the sub-pixel shifting method [35,36], where multiple low-resolution images with sub-pixel shifts were combined to get a high-resolution image. However, the OFM required a close contact distance between the sample and the aperture array in order to achieve high resolution, and the sub-pixel shifting methods need to process multiple low-resolution images which increase the image acquisition time and processing time.

In order to achieve high-resolution imaging with relatively large sensor pixel size in the compact microscopic imaging system, another simple and effective method is to reduce the effective pixel size by interpolation [37–39]. In the case where the spatial frequency of the hologram is less than $1/(2\Delta)$, it may be possible to enhance the resolution by data interpolation because of the effectively oversampling process according to the Nyquist sampling theorem. However, the additional information introduced by the interpolation is an estimation and generally not reliable. In fact, the twin-image background will also affects the resolution of the images, where fine details might be overwhelmed by the twin-image background of the large objects. So in order to obtain high resolution, the twin-image background should also be suppressed as much as possible. And thus the combination of interpolation and iterative phase retrieval would effectively enhance the holographic imaging resolution and improve the SNR and imaging contrast as well. Recently, the propagation phasor approach was proposed to combine phase retrieval and pixel super-resolution and achieve competitive resolution compared to traditional techniques [40]. Nevertheless, the mathematical framework is complicated with some additional assumptions, and generally 8–10 raw measurements were needed.

In this Letter, we propose to use the iterative holographic reconstruction method for phase retrieval in the LIHM with only two intensity measurements as constraints in the iteration and enhance the imaging resolution by data interpolation, which is simple and easy to implement. The iterative phase retrieval algorithm was originally developed by Gerchberg, Saxton (G–S) and Fienup et al. [11,12], where the optical field is iteratively propagated between the imaging sensor and sample planes by numerical computation while constraints in the sensor and sample planes are applied to achieve convergence of the iteration. In our LIHM system, we measured the hologram intensities in two slightly shifted sample planes and did data interpolation to enhance the resolution, then applied the iterative reconstruction algorithm to retrieve the phase of the optical field and successfully reconstruct the enhanced-resolution sample image. Compared to other resolution enhancement technique such as OFM and the sub-pixel shifting techniques, our method provides a simple way to enhance the resolution in a compact lensless microscopic imaging scenario.

2. Methods and analysis

For short sample-to-sensor distance, the diffraction limited resolution calculated by Abbe diffraction is usually less than 1 μm. For example, in the case of our experiment, $\lambda = 473$ nm, $z \sim 5$ mm, $N\Delta \sim 4$ mm we have the finest resolution $R \sim 0.7$ μm according to the Rayleigh criterion.

However, considering the in-line holographic reconstruction for the short sample-to-sensor distance case, we will find that the sampling distance, i.e., the pixel size, limits the resolution. In the reconstruction, the angular spectrum propagation method was one of the most popular techniques to be used, where two fast Fourier transforms can be used to increase the computation time efficiency [41]. In the angular spectrum propagation method, the scalar propagation of the optical field from one plane (plane 1) to another plane (plane 2) can be calculated as

$$U_2(x,y) = FT^{-1}\left\{FT\left\{U_1(x,y)\right\}\cdot\exp\left(i2\pi z\sqrt{\left(\frac{1}{\lambda}\right)^2 - f_x^2 - f_y^2}\right)\right\} \quad (1)$$

where $U_1(x,y)$ and $U_2(x,y)$ are the optical field at plane 1 and 2, respectively, f_x and f_y are the spatial frequencies, z is the propagation distance, and $FT\{\}$ and $FT^{-1}\{\}$ denote the Fourier transform and the inverse Fourier transform, respectively. In digital computation, the discrete Fourier transform need to be applied to the discretely sampled optical field, and after the Fourier transform and the inverse Fourier transform, the sampling distance in plane 1 and plane 2 will be the same. Thus in the in-line holographic reconstruction using Eq. (1), the pixel size in the reconstructed image will be the same as the sensor pixel size. Then according to the Nyquist sampling theorem, the imaging resolution will be 2Δ, which is 4.4 μm in our experiment as we used a CMOS imaging sensor with pixel size of 2.2 μm.

On the other hand, we should notice that the pixel size in the reconstructed image can be smaller if we use other reconstruction methods, such as applying the scalar diffraction formula and calculate the diffraction integral directly [13]. Nevertheless, the sensor pixel size will still be a limiting factor for the imaging resolution because of the sampling requirement. According to the Nyquist sampling requirement, we should have of $1/\Delta \geq 2B_v$, where B_v is the detectable spatial bandwidth of the system and is roughly equal to $N\Delta/2\lambda z$, thus $\Delta \sim 1.1$ μm using the above parameters. Interestingly, the requirement for sampling distance can be lifted for locally band-limited fields according to the generalized sampling theorem [13], where the sampling requirement for the sensor pixel size will be:

$$\frac{1}{\Delta} \geq \frac{B_x}{\lambda z} \quad (2)$$

where B_x is the object size. Thus with similar parameter as above, a sensor pixel size of 2.2 μm would be enough for object size smaller than 1.2 mm. Thus for smaller object size, we should be able to get higher imaging resolution than the Nyquist sampling requirement. This can be achieved by improving the angular spectrum propagation method mentioned above with a simple data interpolation process.

As explained previously, the resolution of LIHM is determined by the sensor pixel size and we should be able to get better resolution for locally band-limited fields. Here we should notice that the resolution enhancement effect depends on the object size and its spatial frequency distribution [42].

To solve the sampling problem in the sample plane while still using Eq. (1) for reconstruction, we propose to decrease the pixel size by data interpolation in the recorded hologram to decrease the pixel size in the reconstructed sample image and thus enhance the resolution. However, the interpolated data is just an estimation of the new data points in the interpolated hologram, which need to be refined for better reconstruction. Furthermore, the direct holographic reconstruction is also disturbed by the twin-image background. To refine the estimated data and eliminate the twin-image disturbance at the same time, the iterative holographic reconstruction method with phase retrieval capability will be an effective approach. We did numerical simulations and experiment to demonstrate the resolution enhancement and image quality improvement by combining the data interpolation with iterative holographic reconstruction method between three planes (the sample plane and the two sensor planes). Here we used the iterative phase retrieval method with two intensity measurements [14] because of its simple experimental implementation. The flow chart of the procedure is presented in Fig. 1, and the procedure in details is as follows:

(1) Interpolate the holograms so that one original pixel will become $4*4$ pixels in the interpolated hologram, as shown in Fig. 2, where A_0 denotes the original pixel and $A_{ij} = A_0/16$ $(i,j = 1\ldots4)$ are the effective pixel values after interpolation. For our simulation and experiment, the pixel size of $A_{ij} = 0.55$ μm. Then initialize the optical fields U_1 (U_2) at z_1 (z_2) with amplitude as the square root of the hologram intensity and phase as zero. Notice that we used the nearest-neighbor interpolation for its simplicity and we observed that similar reconstruction result can also be achieved if linear interpolation was used since the interpolated values would be refined in the iteration process.

(2) Back-propagate U_1 to the sample plane to get U_0, then propagate U_0 to the z_2 distance and get $\hat{U}_2$, and then first let

$$\hat{U}_2(m,n) = \mathrm{abs}(U_2(m,n))\exp\left[i\cdot\mathrm{phase}\{\hat{U}_2(m,n)\}\right] \quad (3)$$