# 2D resolution improvement via 1D scanning Space-Time Digital Holography (STDH) in Optofluidics

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**Abstract.** Space-Time Digital Holography (STDH) exploits the object motion to record the hologram in a hybrid space-time domain. This representation adds new capabilities to conventional DH, such as unlimited extension of the Field of View (FoV) and tunable phase shifting. Here we show that STDH is able to improve the spatial resolution as well. Differently from other super-resolution approaches, stitching between holograms or their spectra is no longer required. Moreover, we introduce a new STDH modality to record and process hybrid space-time representations. This allows improving resolution with one single object scan, paving the way to the use of STDH for superresolution imaging onboard Lab on a Chip devices.

# 1 Introduction

Recently, we introduced a novel recording modality, named Space-Time Digital Holography (STDH), which exploits a projection of the DH fringes to map the hologram signature in a hybrid space-time domain [1-3]. The idea of STDH is to move complexity from the reconstruction to the acquisition process, by imposing a specific constraint on the scanning velocity that has to be matched to the frame rate of the sensor. Thus, a synthetic STDH can be created that possesses all the capabilities of a space-space DH [4,5], along with some added-value features. Above all, whenever the object motion is an intrinsic feature of the system, e.g. in the case of microfluidics, a STDH mapping all the samples flowing inside the channel during a long experiment can be generated and numerically refocused by one single propagation process. Thus, STDH has been shown to be a valuable tool for the analysis with extended FoV of biological samples in Lab-on-a-Chip (LoC) environment, and for high-throughput cell counting and velocimetry by exploiting the sample motion [3]. In this manuscript, we introduce a new STDH modality to record and process hybrid space-time representations. This allows improving resolution with one single object scan. Onion epidermal cells has been used as samples to verify this approach.

## 2 Methods

Take x-direction scanning as an example, when using an  $M \times N$  area array CCD sensor for digital holographic recording of space-time scanning, M columns of pixels can be used for space-time hologram construction. Then, if K frame holograms are recorded, according to the basic space-time hologram composition method, we can build M frame space-time stacks:

$$I_{i}(k,y) = \begin{bmatrix} s_{1,0} & s_{2,0} & \cdots & s_{K,0} \\ s_{1,1} & s_{2,1} & \cdots & s_{K,1} \\ \vdots & & \ddots & \vdots \\ s_{1,N} & s_{2,N} & \cdots & s_{K,N} \end{bmatrix}, (i = 1, \dots, M)$$
(1)

where,  $I_i(k, y)$  (*i*=1,...,M) represent space-time holograms constructed using different columns of pixels. Furthermore, above stacks can be thought of as a series of holograms containing continuously varying phase shift information, like shown below:

$$\begin{cases} I_{0}(k, y) = A(k, y) + 2|E_{0}(k, y)||E_{R}(k, y)|\cos[\varphi(k, y) + \psi_{0}(k, y) + \Delta\psi_{1}] \\ I_{1}(k, y) = A(k, y) + 2|E_{0}(k, y)||E_{R}(k, y)|\cos[\varphi(k, y) + \psi_{0}(k, y) + \Delta\psi_{2}] \\ \vdots \\ I_{M}(k, y) = A(k, y) + 2|E_{0}(k, y)||E_{R}(k, y)|\cos[\varphi(k, y) + \psi_{0}(k, y) + \Delta\psi_{M}] \end{cases}$$

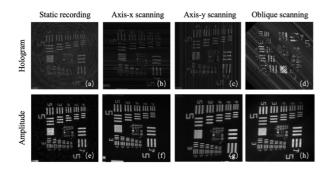
$$(2)$$

where  $E_0$  and  $E_R$  are the object beam and the reference beam, respectively, and  $A = |E_0|^2 + |E_R|^2$  is the zeroth order term. In Eq. (2), STDHs constructed by different sampling columns correspond to the tiny phase shifts,  $\Delta \psi_m$  (m=1,..., M), added to the original information  $\varphi(k, y) + \psi_0(k, y)$ . In this case, the minimum phase shift obtainable is  $\Delta \psi_{min} = (2\pi\Delta x \sin\theta)/p$ , where p is the pitch of the spatial carrier, and  $\theta$  is the angle between the fringe direction and the x-axis. If four proper columns, j, k, l, m, are selected, STDHs with  $\Delta \psi_{j,k,l,m} =$  $[0, \pi/2, \pi, 3\pi/2]$  are obtainable, which is sufficient to apply a four-step phase shifting formula as in [1]. Then, numerical refocusing can be applied.

#### **3 Experimental results**

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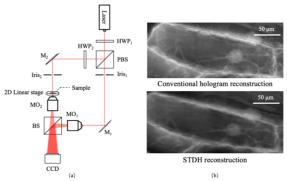
We use a banded area of a CCD sensor to capture a set of observables while the object moves. For each object position, a column (or row) of pixels, which records a 1D signal, is acquired in the form of modulated fringes. Moreover, we use a diagonal pixel set to acquire 2D signals, which is an efficient way to scan that we recently introduced [6]. The acquired sequence is then used to synthetize the STDH that corresponds to the modulation of a new carrier. The idea of STDH is to move complexity from the reconstruction to the data capture process, by imposing a specific constraint on the scanning velocity that has to be matched to the frame rate of the sensor. Thus, a synthetic STDH can be created maintaining all the features of a space-space DH, along with some novel ones.



**Fig. 1.** Preliminary experimental results of self-assembling modality. (a) conventional hologram; (b)-(d) space-time holograms by x, y and oblique scanning; (e)-(h) amplitude reconstruction results.

To verify the resolution improvement process of selfassembling modality in STDH, a series of experiments were performed by using an USAF-1951 resolution target, as shown in Fig. 1. When the sample was scanned in 1D direction (x-axis or y-axis), the resolution under scanning direction could be improved by self-assembling of object frequencies [6]. Furthermore, once the sample was scanned in oblique direction, i.e. introducing velocity components both in x direction and y direction, the resolution of axis-x and axis-y could be improved at the same time. In summary, comparing STDH to conventional DH imaging, we notice that the object frequencies self-assembly process of space-time scanning can effectively improve the imaging resolution. Moreover, the Signal-to-Noise Ratio (SNR) of STDH is higher than the SNR of a conventional DH reconstruction, without requiring any further denoising process [7].

For onion cells, the holographic recording system is based on an off-axis Mach-Zehnder interferometer with coherent light source which wavelength is 532.8nm. The laser beam is divided into two beams by a polarizer beam splitter prism after simple filtering. The onion epidermal cells are placed on a 2D linear stage, it is used to achieve x-y plane scanning for STDH recording. Two 25× objective lenses are used for magnification imaging. A scientific-grade CCD camera is placed after beam splitter prism, the pixel size is  $4.5 \mu m \times 4.5 \mu m$ , and the screen size is  $1280 \times 960$ .



**Fig. 2.** STDH recording and reconstruction of onion epidermal cells. (a) experimental setup; (b) phase reconstruction results, the upper image is from conventional hologram, the lower image is from STDH.

As shown in Fig. 2(b), the phase image of STDH reconstruction results exhibits a clear resolution increase, with respect to conventional hologram reconstruction result. The internal structure of the onion epidermal cells can be observed more clearly. This is a further evidence that the self-assembly process improves resolution in 2D by 1D scanning imaging.

## 4 Conclusions

Thanks to the self-assembling of object frequencies, the resolution of Space-Time Digital Holography has been improved. In the preliminary experiment, we verified the improvement of imaging resolution by different scanning directions and proved that the imaging resolution in X and Y directions can be improved simultaneously in the oblique recording process. Also, in the experiment for onion epidermal cells, self-assembly process in STDH approach provided more detailed information in phase image with respect to conventional digital holography.

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