Electronic Supplementary Material (ESI) for Lab on a Chip.

Supplementary Information

Muscope: a miniature on-chip lensless microscope

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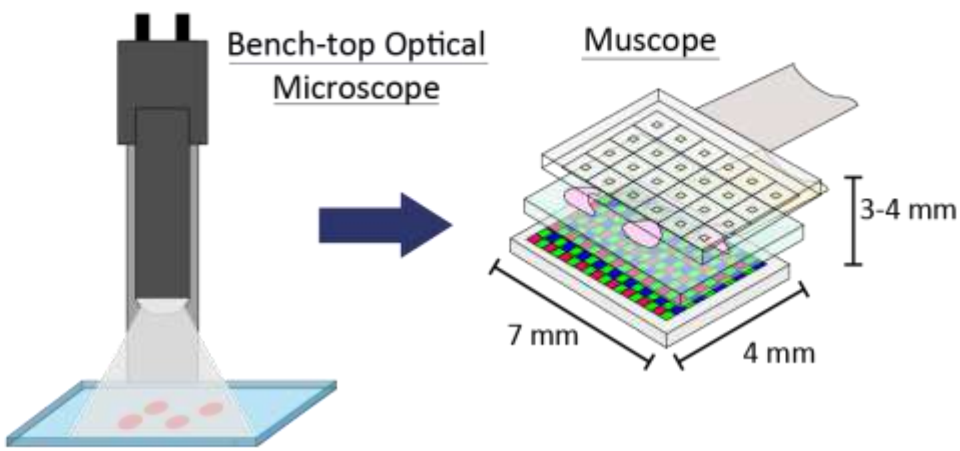

Contents

1. Holographic Reconstruction Algorithm

For the imaging setup shown in Fig.1(f), microLEDs were considered as point emitters. Thus the object plane receives spherical wave

$$U_{incident} = exp(ikr)/r$$

here, $r = (\boldsymbol{x}, \boldsymbol{y}, \boldsymbol{z})$ and z is the imaging axis. Under the assumption that the Fresnel's number $N_f = n(object\ size)^2/\lambda z_2$ is small, it can be shown that the hologram captured by the sensor is given by eq.1,[1]

$$H(X, Y) = |t(X, Y) \otimes s(X, Y)|^2 \qquad (1)$$

where $R = (X, Y, Z)$ are coordinates in sensor plane, $t(x, y)$ is the transmission function of the object, $\otimes$ is the convolution operator and Fresnel's function

$$S(u, v) = exp(-i\pi\lambda z(u^2 + v^2))$$

acts as a propagator and. The hologram also gets magnified by a factor

$$M = (z_1 + z_2)/z_1.$$

A reconstruction procedure propagates the intensity pattern $H(X, Y)$ back to the object plane to find the transmission function of the object. The absence of phase information from the hologram makes this process imperfect, in particular, leading to twin images. An interferometric phase retrieval algorithm[2] deals with these problems and was used here for image reconstruction. The algorithm works well for near field holograms which have Fresnel's number N_f ~ 1 or lower. For

Muscope setup, this condition is satisfied readily with objects of size 10 μm or lower. In near field imaging, the intensities are dominated by diffraction terms and the real and virtual images are not well separated out. The iterative algorithm clips one of the twin images in appropriate planes to reduce the effect of the other. The algorithm first generates the virtual image from the intensity of the captured hologram by forward propagating it to a distance z_2. The object support is detected in this reconstructed image and is replaced by the average of the reconstructed image inside the support. The reconstruction is then back propagated to the object plane (-2z_2). A gradual clip operator is applied to the generated image to set the field outside the support equal to background. The resulting image is forward propagated to the virtual image plane again (2z_2). The signal inside the support is again replaced by the average calculated in initial pass. The image is again back propagated to the object plane to get a reconstruction. The process converges after several iterations. For our reconstructions, 5-10 iterations were sufficient. As noted above, the forward and backward propagation used spherical waves under Fresnel-Kirchoff approximation. The value of z_2 which yielded sharpest images was used for reconstruction. The refractive index of the medium was a linearly weighted sum of refractive indices of glass (1.33) and air (1.0), where the weights were the fraction of z_2 covered by each. Two coverslips in sample to sensor path resulted in glass thickness of 340 μm.

2. Super-resolution Algorithm

Muscope supports programmable movement of the light source that can be used to implement several super-resolution techniques available in the literature.[3-4] The resolution of the reconstructed images depends on several factors. These factors include spatially incoherent and wideband emission from the microLEDs, finite sensor pixel dimensions, low signal to noise ratio (SNR) and approximations made in the reconstruction algorithm. The SNR is affected by the sensitivity of the sensor, the absorbance of the Bayer layer, the illumination intensity from the microLEDs and several noise sources. A combination of these factors limits the number of fringes in the hologram of an object. The outermost fringes contain the high frequency components of the hologram and therefore, the finer features of the sample. Even with highly coherent and high intensity lasers, the fringes which are only as fine as a multiple of the sensor pixel size can be analysed (Nyquist criterion).

The impact of finite pixel size of the sensor can be reduced by a multi-image super-resolution algorithm, FastSR.[4] It fuses several low resolution but subpixel shifted images to make a high resolution one. With Muscope, the low resolution images were obtained by shifting the position of the microLED on the display array. The change in position of microLED caused a shift in the holograms on the image sensor as shown in Fig.3(a). With the knowledge of the size of the sensor pixel, the pitch of the microLED display and the distances involved, one can select illumination to result in known (sub-) pixel shifts in holograms. To obtain a N times higher resolution image, the subpixel shifts were binned into bins of size 1/N, in both directions, with a total of N^2 bins within a pixel. For each pixel in low resolution images there were N^2 pixels in the high resolution image, contributed by these N^2 bins. Normally, an illumination pattern will lead to bins having no images or bins having more than one image. In the latter case, median values of all the images in that bin are used, which is a robust estimate for the pixel value. In the former case, an interpolation scheme is needed.

The shift between a pair of holograms from neighboring illuminated microLEDs was determined by the algorithm of Feinup et al.[5] The algorithm allows detection of sub-pixel shifts located in bins, whose size can be controlled by passing the upsample factor N to the algorithm.

In the second step of the super-resolution algorithm, the effect of the point spread function of the sensor pixels is removed by a gradient descent scheme based on a L1 norm. The presence of noise and the need for interpolation is also taken into account in this step. The point spread function of the image sensor has been determined using method by Ozcan et al.[6]

References

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