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

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Optofluidic ptychography on a chip

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Supplementary Note 1: Tracking positional shift and rotation angle

In order to recover the positional shift and rotation angle via correlation analysis. We first average all measurements and get a rough estimate of the scattering layer. The captured raw images are divided by this estimate to remove the modulation pattern on the raw images. We then digitally propagate the processed raw images to the plane of the microfluidic channel like that in digital in-line holography. Finally, the positional shift and the rotation angle are recovered using the following optimization process:

$$(\hat{x}_i, \hat{y}_i, \hat{\theta}_i) = \underset{x_i, y_i, \theta_i}{\arg min} \sum_{(x,y)\in C} \left[T_{x_i, y_i, \theta_i}\{O_1(x,y)\} - O_i(x,y)\right]^2 \quad (1)$$

where $O_i(x, y)$ is the digital in-line holographic reconstruction for the i[th] captured image. *'C'* donates the region of the tracked object. x_i, y_i and θ_i in the subscript of $T\{\cdot\}$ represent horizontal shift of x_i, vertical shift of y_i, and θ_i rotation. $T\{\cdot\}$ is a transform operation of an image for positional shift and rotation, which can be expressed as

$$\begin{aligned} T_{x_i, y_i, \theta_i}\{O_1(x,y)\} &= O_1(x\cos\theta_i - y\sin\theta_i + x_i, y\cos\theta_i + x\sin\theta_i + y_i) \\ &\approx O_1(x - y\theta_i + x_i, y + x\theta_i + y_i) \\ &\approx O_1(x,y) + (x_i - y\theta_i) g_x(x,y) + (y_i + x\theta_i) g_y(x,y), \end{aligned} \quad (2)$$

where $g_x(x,y) = \frac{\partial O_1(x,y)}{\partial x}$ and $g_y(x,y) = \frac{\partial O_1(x,y)}{\partial y}$. The first approximations will be satisfied when θ_i has a small value and we adopt the first three terms of the Taylor series expansion in the second approximation. With Eq. (2), the Eq. (1) can be rewritten as

$$\begin{aligned} (\hat{x}_i, \hat{y}_i, \hat{\theta}_i) &= \underset{x_i, y_i, \theta_i}{\arg min} \sum_{(x,y)\in C} \left[O_1(x,y) + (x_i - y\theta_i) g_x(x,y) + (y_i + x\theta_i) g_y(x,y) - O_i(x,y)\right]^2 \\ &= \underset{x_i, y_i, \theta_i}{\arg min} E_i(x_i, y_i, \theta_i) \end{aligned} \quad (3)$$

By differentiating $E_i(x_i, y_i, \theta_i)$ with respect to x_i, y_i, θ_i and then set the derivatives equal to zero, we can obtain the $\hat{x}_i, \hat{y}_i, \hat{\theta}_i$ to minimize Eq. (1). The expression of estimated registration vector can be computed as

$$(\hat{x}_i, \hat{y}_i, \hat{\theta}_i)^T = M^{-1}N, \quad (4)$$

where

$$M = \begin{bmatrix} \sum {g_x}^2(x,y) & \sum g_x(x,y)g_y(x,y) & \sum \overline{g(x,y)}g_x(x,y) \\ \sum g_x(x,y)g_y(x,y) & \sum {g_y}^2(x,y) & \sum \overline{g(x,y)}g_y(x,y) \\ \sum \overline{g(x,y)}\, g_x(x,y) & \sum \overline{g(x,y)}\, g_y(x,y) & \sum \overline{g(x,y)}^2 \end{bmatrix} \tag{5}$$

$$N = \begin{bmatrix} \sum f(x,y)g_x(x,y) \\ \sum f(x,y)g_y(x,y) \\ \sum f(x,y)\, \overline{g(x,y)} \end{bmatrix} \tag{6}$$

$$\overline{g(x,y)} = xg_y(x,y) - yg_x(x,y) \tag{7}$$

$$f(x,y) = O_i(x,y) - O_1(x,y) \tag{8}$$

Supplementary Note 2: Image rotation via shearing operations in the Fourier domain

To perform a rotation of an image, we use an algorithm based on the decomposition of the 2D rotation into the product of three shear matrices. Considering a general 2D clockwise rotation by an angle θ as follows

$$\begin{pmatrix} x' \\ y' \end{pmatrix} = \begin{pmatrix} \cos\theta & \sin\theta \\ -\sin\theta & \cos\theta \end{pmatrix} \cdot \begin{pmatrix} x \\ y \end{pmatrix}$$

$$= \begin{pmatrix} 1 & \tan\frac{\theta}{2} \\ 0 & 1 \end{pmatrix} \cdot \begin{pmatrix} 1 & 0 \\ -\sin\theta & 1 \end{pmatrix} \cdot \begin{pmatrix} 1 & \tan\frac{\theta}{2} \\ 0 & 1 \end{pmatrix} \cdot \begin{pmatrix} x \\ y \end{pmatrix} = M_x \cdot M_y \cdot M_x \cdot \begin{pmatrix} x \\ y \end{pmatrix} \qquad (9)$$

Here, (x, y) are the original image coordinates. The new image coordinates are (x', y') where the image has been rotated clockwise by an angle θ. M_x and M_y are the shearing matrices along the x-direction and y-direction. From Eq. (9), we can see that a rotation matrix can be decomposed into three simple shearing transformations (x-direction shear, then y-direction shear, x-direction shear again). Next, we exploit the relationship between the shearing transformation and the Fourier shift theorem. Assuming an original image $h(x, y)$, the image after the first x-shearing operation can be expressed as

$$g_r(x', y') = h\left(x + \tan\frac{\theta}{2} \cdot y, y\right) \qquad (10)$$

We can then apply 1-dimension Fourier transform (1D FT) along the direction of shear

$$G_r(u, y') = \exp(-2\pi iuay)\, H_r(u, y) = \exp\left(-2\pi iu \tan\frac{\theta}{2} y\right) \mathcal{F}_r\{h(x, y)\} \qquad (11)$$

Here the subscript '*r*' means the shearing transformation along the x-direction (rows). $G_r(u, y')$ and $H_r(u, y)$ are the 1D FT of $g_r(x', y')$ and $h(x, y)$ along the rows. $\mathcal{F}_r\{\cdot\}$ represents 1D FT. According to Eqs. (10-11), an x-sheared operation can be performed as

$$g_r(x', y') = \mathcal{F}_r^{-1}\left\{\exp\left(-2\pi iu \tan\frac{\theta}{2} y\right) \mathcal{F}_r\{h(x, y)\}\right\} \qquad (12)$$

Similarly, we can perform a y-shear and obtain:

$$g_{cr}(x', y') = \mathcal{F}_c^{-1}\{\exp(-2\pi iv(-\sin\theta)x)\, \mathcal{F}_c\{g_r(x', y')\}\} \qquad (13)$$

The subscript '*c*' means the shearing transformation along the y-direction (columns). $\mathcal{F}_r\{\cdot\}$ represents 1D FT along the y-direction (columns). Finally, we perform an x-shear again

$$g(x', y') = g_{rcr}(x', y') = \mathcal{F}_r^{-1}\left\{\exp\left(-2\pi iu \tan\frac{\theta}{2} y\right) \mathcal{F}_r\{g_{cr}(x', y')\}\right\} \qquad (14)$$

We obtain an image $g(x', y')$ after rotating $h(x, y)$ by an angle θ. The above procedure defines the three-step (shearing transformation) process of a rotation. That is to say a θ rotation can be accomplished by three 1D forward FTs and three 1D inverse FTs performed on 2D images.

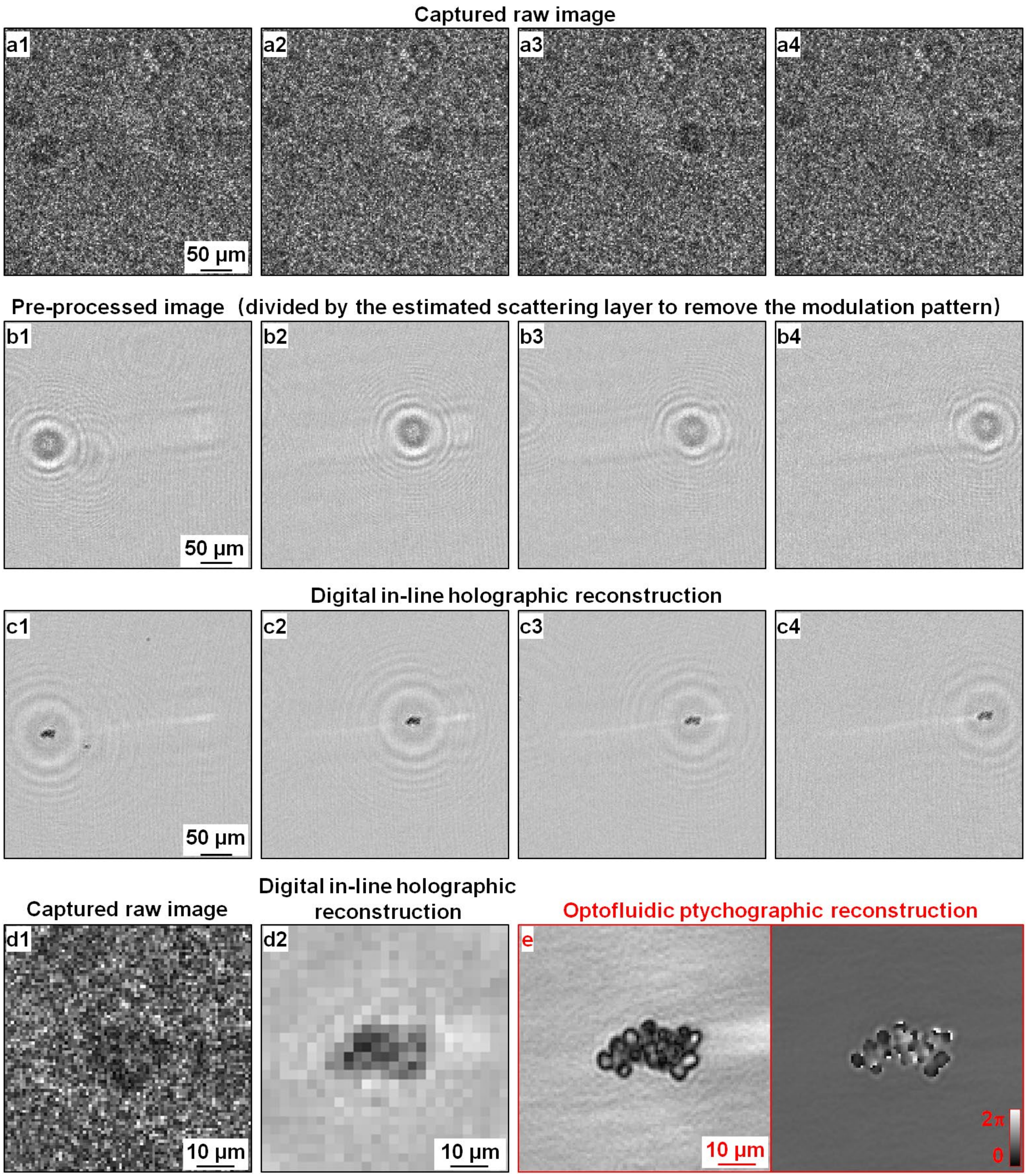

Fig. S1: Image processing for estimating positional shifts and rotation. (a) The captured raw images of yeast cells. (b) Pre-processed images, where each raw image is divided by the estimated scattering layer to remove the modulation pattern. (c) Digital in-line ptychographic reconstruction. (d1-d2) Magnified views of (a1) and (c1). (e) Optofluidic ptychographic reconstruction of intensity and phase.

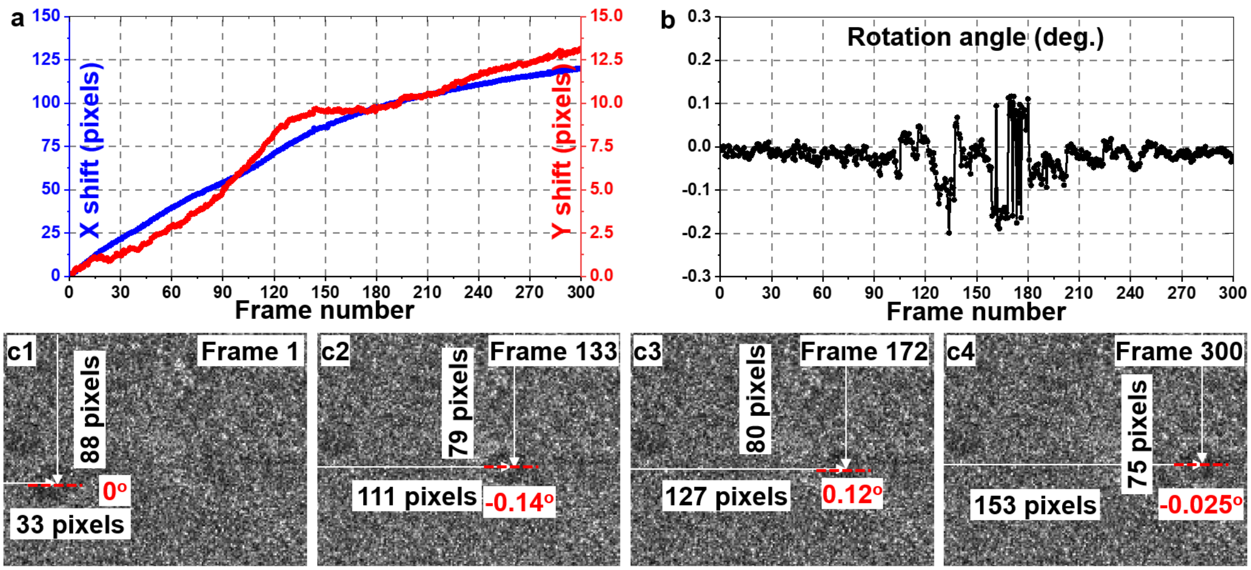

Fig. S2: Tracking the positional shifts and the rotation angle of objects passing through the channel. (a) The tracked positional shifts of a cluster of yeast passing through the channel via electrokinetic flow. (b) The tracked rotation angle of the yeast. (c) The raw images are processed to generate digital in-line holographic reconstruction for the tracking process.

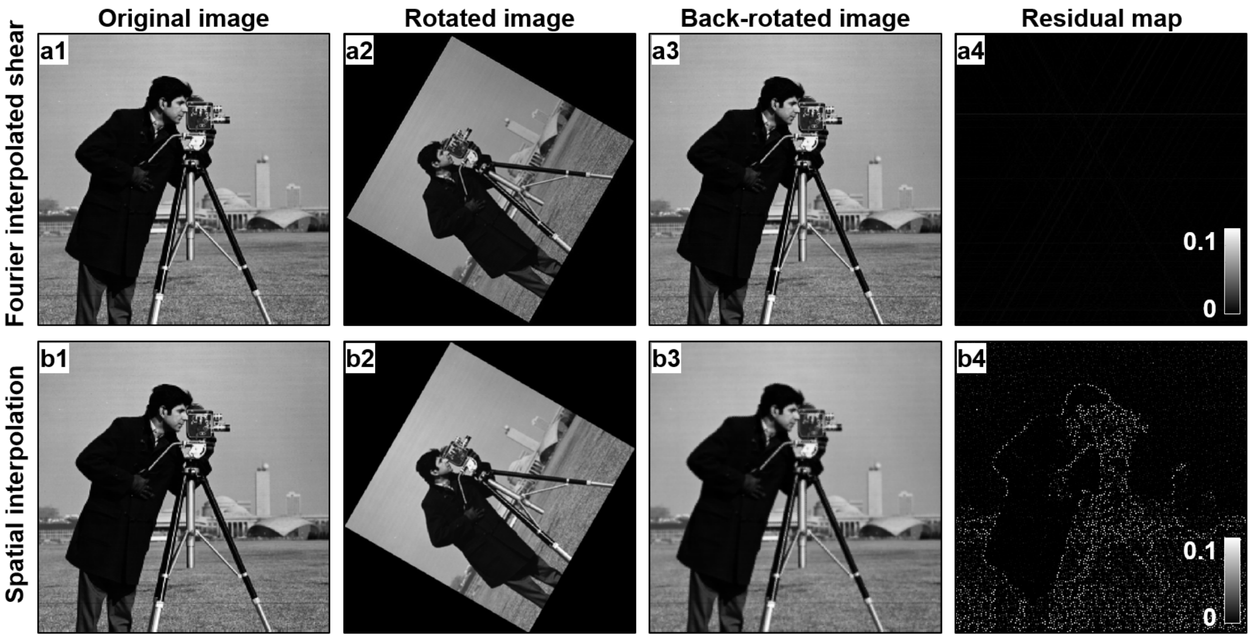

Fig. S3: Rotation via Fourier shearing and spatial interpolation. (a1-b1) Original images. (a2-b2) Images by rotating the original image by 30°. (a3-b3) Back-rotated images. (a4-b4) Residual map between the original image and the back-rotated image of the two applied methods. We can see that the regular spatial interpolation method is not reversible.

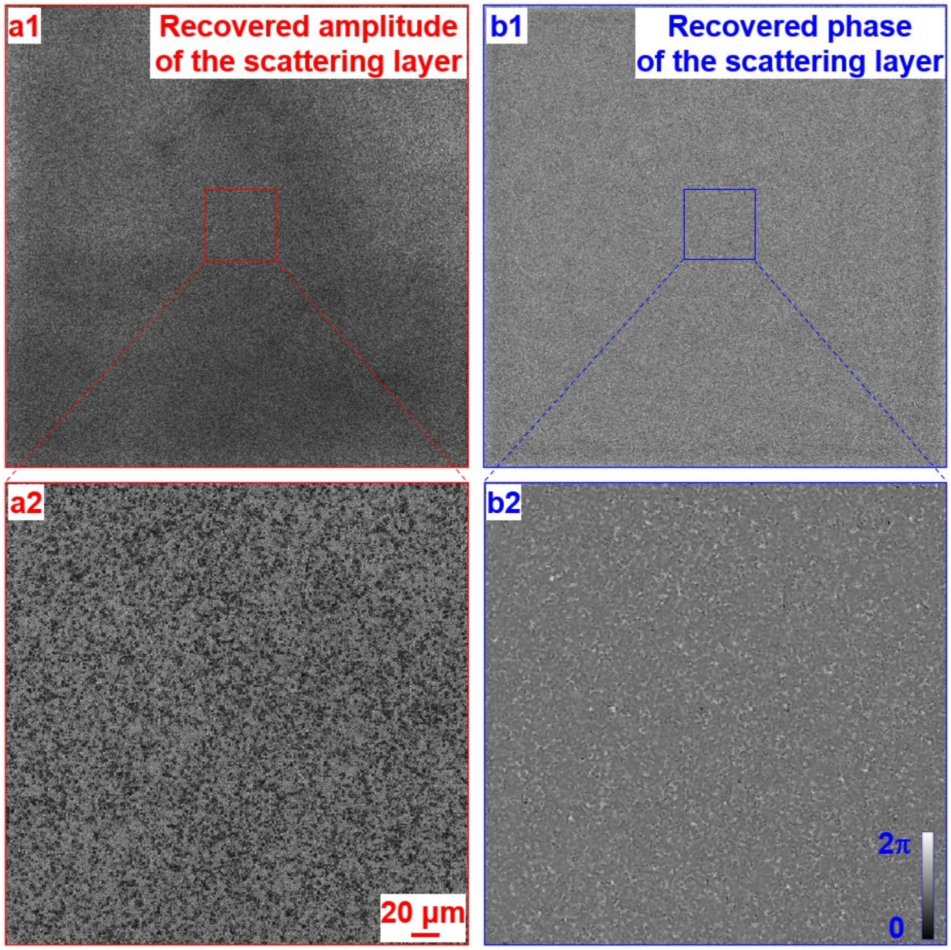

Fig. S4: The recovered amplitude (a) and phase (b) of the scattering layer.

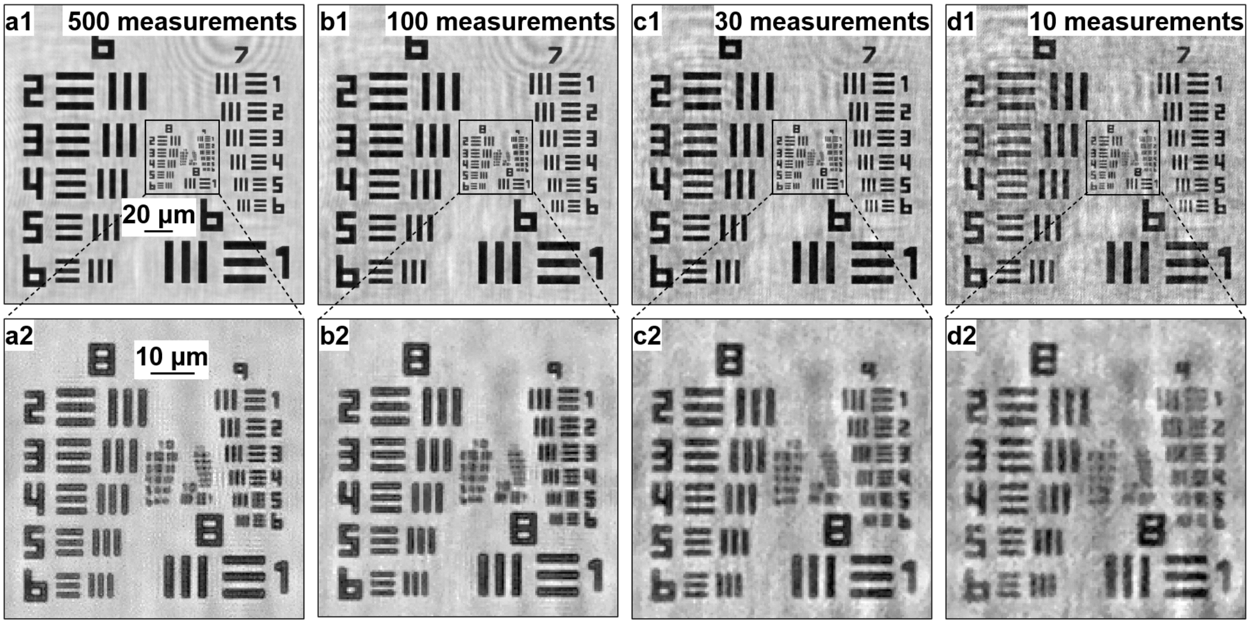

Fig. S5: Optofluidic ptychographic reconstruction using different numbers of raw images. (a) 500 images. (b) 100 images. (c) 30 images. (d) 10 images.