Wavefront Analysis and Optimization from Conventional Liquid Crystal Displays for Low-Cost Holographic Optical Tweezers and Digital Holographic Microscopy

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ABSTRACT
In different study fields the manipulation and imaging of micro-sized particles is essential. The use of holographic optical tweezers (HOT) and digital holographic microscopy (DHM) facilitates this task in a non-mechanical way by providing the proper computer generated hologram and the required amount of light. Electrically addressed spatial light modulators (EASLM) found in holographic optical tweezers are typically of the reflective liquid crystal on silicon (LCoS) type which can achieve a phase shift of more than $2\pi$ but they are expensive. Similar components like transmissive twisted nematic liquid crystal displays (TN-LCD) are produced in large quantities, their optical characteristics improve rapidly and they are inexpensive. Under certain circumstances these devices can be used instead of expensive spatial light modulators.

Consumer grade objectives are not always well corrected for spherical aberration. In that case conventional liquid crystal displays can also compensate these undesired optical effects. For this purpose software-corrected computer generated holograms are calculated. Procedures to analyze and compensate different parameters of a conventional low-cost liquid crystal display, e.g. phase shift evaluation and aberration correction of objectives by Zernike polynomials approximation are explained. The applied software compensation of the computer generated hologram has shown significant improvement of the focus quality. An important price reduction of holographic devices could be achieved by replacing special optical elements if correction algorithms for conventional liquid crystal displays are provided.

Keywords: Wavefront Correction, Holographic Optical Tweezers, Spatial Light Modulators, Digital Holographic Microscopy

1. INTRODUCTION
For certain tasks in microscopy research it is necessary to manipulate small particles. Optical tweezer instruments use the attractive forces of a tightly focused laser beam to dielectric particles. For example polystyrene beads in water with a diameter of 2.2 $\mu$m which have a higher refractive index than the surrounding medium will experience an attractive force in the direction of the gradient of light intensity. These forces (typically some piconewtons$^1$) can be used to trap and move particles in three dimensions by moving the ambient chamber or deflecting the laser beam. An outline of these forces is shown in Fig. 1 left (adapted from Williams 2002). Both procedures represent a drawback since it is difficult and expensive to translate optical elements with a resolution and repeatability of $<1 \mu$m. Also the accelerating masses could introduce vibrations, which makes it necessary to wait until the vibration decays. Because the perpendicular radiation pressure pushes the particle away$^{1,2}$ from the beam waist the gradient and total power must be high and therefore the numerical aperture has to be sufficient large. Also beam distortions from aberration will reduce the trapping forces.

HOT can create and move single or multiple foci by manipulating the incoming wavefront with EASLM so that the interfering monochromatic light forms the traps. Because the trapping forces of HOTs are based on the same physical principle as conventional optical tweezers they also depend on a beam waist with high gradient

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Figure 1. Schematic diagram showing forces $F_{\text{refl}}$ and $F_{\text{refr}}$ on a dielectric sphere due to both reflection and refraction of two rays of light (left) and schematic of a HOT setup (right).

Figure 2. Schematic diagram of a DHM in transmissive setup with spatial light modulator which creates the illumination and reference wave or special shaping to archive high trapping forces. The use of phase modulating EASLMs suggests itself that these are not only used to holographically create the traps but also to correct some of the aberrations which are introduced from non-ideal optical elements.

DHM can be distinct from other microscopy methods by not recording an image formed by a lens. Instead, the interference pattern from the object wave and a reference wave form a hologram which is recorded and adjacent the object is reconstructed with numerical algorithms. Figure 2 shows the used setup. A hologram on the EASLM creates a wavefront which forms two foci at different distances. One illuminates the object and the other is used as reference wave. The phasing between these waves can be controlled by the SLM and is used to take five pictures with different illumination and phasing on the CMOS camera. Since only the hologram is changed on the SLM there is no mechanical movement which could distort the image due to vibrations. The complex object wave can then be reconstructed and slices through the object are calculated offline. This reconstruction process requires a known wavefront for illumination and reference wave which can be characterized with mathematical models. Wavefront distortions such as spherical aberrations or coma which were not considered will decrease image quality and resolution.

For this reason it is important for both the holographic optical tweezers and the digital holographic microscope with spatial light modulator to create sharp and undistorted foci and both applications need objectives
with short focal length and high numerical apertures with low wavefront distortion. Microscope objectives are well corrected for spherical aberration and distortions introduced from coverslip etc. However in order to get large light intensities we used a fast camera lens (Carl Zeiss Planar T* 1.4/50) instead of a microscope objective. But the camera lens is calculated for working distances from 0.5 m to infinity. So fast objectives matching the LCD dimension and optimized wavefront are special parts and therefore expensive. The next chapter will outline the used setup to measure some distortions and calculate a correction which can be applied to the LCD.

2. PROPOSED APPROACHES

2.1 Phaseshift Measurements

The 1.3” p-Si AMLCD (active matrix LCD) used originates from a NEC MT1030G+ beamer and features 1024x768 pixel (width ≈ 26 mm, height ≈ 20 mm) which results in a quadratic pixel size of 25.8 μm. Given that this LCD is normally used to modulate the amplitude of light it is placed between two polarizers, one directly glued onto the LCD. This foil polarizer must be carefully removed with scalpel and tweezer and the remaining adhesive washed off with solvent. The LCD can then be controlled with a resolution of 8 bits per channel (0 :: 2^8 − 1 = 255) via VGA interface.

A Mach-Zehnder interferometer was build to measure the LCD phase shift as a function of the set gray value. Figure 3 shows the used setup. A linear polarized He-Ne laser beam (λ = 632.8 nm) is coupled into a single mode optical fiber and enters a polarization controller. The bended fiber facilitates stress introduced birefringence so that the state of polarization of the Gaussian beam emitting at the fiber end can be controlled by twisting the paddles. An aperture in front of the LCD has two drilled holes so that the light passing each hole illuminates one side of the LCD respectively. One side of the LCD is set to the maximum gray value (2^8 − 1 = 255) and the other side is increased from 0 to 255. This introduces a phase shift to one path of the Mach-Zehnder interferometer. These two waves interfere and the resulting interference pattern past the lens is magnified with a microscope objective and recorded with a CMOS camera. Figure 4 shows one of the recorded pictures.

The phase shift in one path of the interferometer was calculated from the interference pattern after each change of the gray value. This was accomplished by averaging over multiple vertical lines and adjacent fast Fourier transform (FFT) of the resulting horizontal vector into a spectrum. After removing the average, the maximum in the spectrum was found and the phase and amplitude information were extracted. Figure 5a shows the calculated phase and visibility of the interference pattern.

To achieve the maximum possible phase shift and visibility the input polarization state must be adjusted with the fiber polarization controller. Best results were archived with an input polarization (Jones vector)
Figure 4. Inverted (black represents high intensity) interference pattern recorded from the Mach-Zehnder interferometer together with the horizontal area which is used to calculate the phase (thin vertical line).

(a) Dependency of gray value control signal to phase shift (lower) and interferometric visibility (upper) of the recorded interference pattern.

(b) Approximated phase shift to gray value (Eq. 1, \( \nu = 3.4 \text{ rad} \)).

Figure 5. Determined and approximated phase shift for used TN-LCD.
$J = [1; 1.7 \cdot e^{1.1}]$. Furthermore brightness and contrast on the MT1030G+ were set to 50% (value selected from experimental results in the range from 10% to 90%).

The desired achievable phase shift would be $2\pi$ but the measurement had shown that the used TN-LCD can shift the phase at most for $\approx 3.4$ rad. Therefore an approximation has to be done which is achieved by a mapping function which assigns fixed gray values to phases which can not be accomplished. See also Fig. 5b

$$
\Theta(\phi, \nu) = \begin{cases} 
0 & \phi \cdot \frac{255}{\nu} > 255 \wedge \phi > \frac{(2\pi - \nu)}{2} + \nu \\
255 & \phi \cdot \frac{255}{\nu} > 255 \wedge \phi \leq \frac{(2\pi - \nu)}{2} + \nu \\
\phi \cdot \frac{255}{\nu} & \text{else}
\end{cases} \tag{1}
$$

$\Theta$ VGA gray value 0..255

$\phi$ desired phase shift

$\nu$ max. achievable phase shift $\approx 3.4$ rad with used TN-LCD

Target values for phase shift between 0 and $\nu$ can be assigned proportional to gray values from 0 to 255. If the target value exceeds $\nu$ the gray value is set to 255 until the desired target value is closer to $2\pi$ than to $\nu$.

### 2.2 Aberration Measurements

The wavefront distortion in the presented DHM setup, which is mainly introduced from the Zeiss Planar T* objective and warping of the LCD, can be measured with a scanning algorithm on the LCD with a similar setup as Fig. 3 but with removed aperture. We have examined several approaches to retrieve them from the CMOS image data and minimize their effects by superposing a calculated correction function. First we define some functions which will be used below.

The Hadamard product, also known as Schur product if formally

$$
A \in \mathbb{R}^{m \times n} \quad B \in \mathbb{R}^{m \times n}
$$

$$
(A \odot B) \in \mathbb{R}^{m \times n}
$$

with elements $A \odot B$ given by

$$(A \odot B)_{i,j} = A_{i,j} \cdot B_{i,j}$$

Digital aperture functions, for example the circular aperture, are used to minimize the effective angle of beam or to mask parts of the hologram.

$$
A_C \in \{0;1\}^{h \times w}
$$

$$
A_C(x_c, y_c, r) = \begin{bmatrix}
\sqrt{(-\frac{w}{2} - x_c + i - 1)^2 + (-\frac{h}{2} - y_c + j - 1)^2} < r \\
\sqrt{(-\frac{w}{2} - x_c)^2 + (-\frac{h}{2} - y_c)^2} < r & \cdots & \sqrt{((\frac{w}{2} - 1) - x_c)^2 + ((\frac{h}{2} - 1) - y_c)^2} < r \\
\vdots & \vdots & \vdots \\
\sqrt{(-\frac{w}{2} - x_c)^2 + ((\frac{h}{2} - 1) - y_c)^2} < r & \cdots & \sqrt{((\frac{w}{2} - 1) - x_c)^2 + ((\frac{h}{2} - 1) - y_c)^2} < r
\end{bmatrix} \tag{3}
$$

$x_c, y_c$ center position of circular aperture [px]

$r$ radius of circular aperture [px]

$w, h$ number of horizontal and vertical pixel of calculated aperture
Calculated Fresnel lens for phase modulating SLM

\[
f_L(x, y, x_c, y_c, f) = \sqrt{(x - x_c)^2 + (y - y_c)^2 + f^2} \cdot k
\]

\[
u(\phi) = \phi - \left\lfloor \frac{\phi}{2\pi} \right\rfloor \cdot 2\pi
\]

\[
F_L(x_c, y_c, f, \phi_o) = \left[ \begin{array}{c}
u(f_L(-\frac{w}{2}, -\frac{h}{2}, x_c, y_c, f) + \phi_o) \\
\vdots \\
u(f_L(-\frac{w}{2}, \frac{h}{2} - 1, x_c, y_c, f) + \phi_o)
\end{array} \right]
\]

*PS* size of quadratic pixel of LCD [m]

*k* wavenumber \(\frac{2\pi}{\lambda}\)

*x_c, y_c* center of Fresnel lens [px]

*f* focal length [m]

*\(\phi_o\)* phase offset [rad]

In order to determine aberrations, a calculated Fresnel lens \(F_L\) with a focal length \(f = 1.5\, \text{m}\) is displayed on the LCD and the resulting spot is magnified with a X20 objective and captured with a CMOS camera. Since the X20 objective is mounted in the designed position (160 mm length) aberration introduced from this can be neglected. By modifying \(A_C\) it is possible to use only a circular section of the Fresnel lens \(F_L\). The surrounding area is set to a fixed value and contributes to the zero order hologram.

\[
LCD_{C}(\ldots, \phi_o, r_A) = \Theta(F_L(x_{cB}, y_{cB}, f_B, 0) \odot A_C(x_{cA}, y_{cA}, r_A))
\]

If the zero order is not blocked it interferes with the generated focus and forms an interference pattern which is shown in Fig. 6. This makes it difficult to detect the center of the generated focus without some enhanced feature detection. For this reason we decided to block the zero order with a thin melted copper wire which was fixed to a 3D translation stage.

One approach is to set \(A_C(x_c, y_c, r)\) to an arbitrary position and change the slope of the 2D correction function iteratively until the center of intensity lies on the optical axis. This works but the resolution for higher order aberrations is reverse proportional to the diameter of \(A_K\). Therefore decreasing \(r\) would enhance the resolution.
but decreases the amount of light, which contributes to the focus, and the numerical aperture which increases the beam waist. Furthermore this iterative scanning is very time-consuming and in addition hard to integrate the several X,Y slopes into one consistent model.

The other approach is to use a centered annulus aperture

\[ A_A(r_1, r_2) = A_K(0, 0, r_1) \land A_K(0, 0, r_2) \]

(9)

and adapt the focal length \( f_B \) and offset \( \phi_0 \) in an approximation procedure so that the intensity on the optical axis gets maximized. Experiments have shown that this only works for aberrations which are concentric around the optical axis, which is not the case in typical systems.

A successful approach was to scan the hole system with the hologram

\[ LCD_C(x_cB = 0, y_cB = 0, x_cA, y_cA; f_B) \]

(11)

(see also Eq. 8) by increasing \( x_cA \) from \( -\frac{w_2}{2} \) to \( \frac{w_2}{2} \) and \( y_cA \) from \( -\frac{h_2}{2} \) to \( \frac{h_2}{2} \) in steps of \( \Delta s \). After each step a picture of the intensity in the image plane is taken with the CMOS camera and the center of intensity \( C_I(n) \) is calculated. Subsequent the position \( x_cA(n), y_cA(n) \) and radius \( r_A \) of the aperture, the center of intensity \( C_I(n) \) and the average intensity is stored to a file for post processing.

After calculating a correction function by approximation with Zernike polynomials (see next chapter 2.3) the X20 objective is replaced with a X40 microscope objective to increase the resolution in \( C_I \) and the hole procedure is run again with the correction function from the first run applied to the hologram.

### 2.3 Zernike Polynomials

The even Zernike polynomials are defined as

\[ Z_n^m(\rho, \phi) = R_n^m(\rho) \cos(m \phi) \]

(12)

and the odd ones as

\[ Z_n^{-m}(\rho, \phi) = R_n^m(\rho) \sin(m \phi), \]

(13)

where \( m \) and \( n \) are nonnegative integers with \( n \geq m \). \( \phi \) is the azimuthal angle and \( \rho \) is the radial distance. The radial polynomials \( R_n^m \) are defined as

\[
R_n^m(\rho) = \begin{cases} 
\sum_{k=0}^{(n-m)/2} (-1)^k \binom{n}{k} \frac{(n-m)!}{k!(n-k)!} \rho^{n-2k} & \text{n-m even} \\
0 & \text{n-m odd}
\end{cases}
\]

(14)

We used Zernike polynomials to approximate our collected distortions (see chapter 2.2) up to order 8 which leads to a 45 column vector

\[
Z_{\text{reg}} \in \mathbb{R}^{45 \times 1}
\]

(15)

\[
Z_{\text{reg}}(r, \phi) = \begin{bmatrix} 1 \\ r \cdot \cos(\phi) \\ r \cdot \sin(\phi) \\ r^2 \cdot 2 - 1 \\ r^2 \cdot \cos(2 \cdot \phi) \\ \vdots \\ r^8 \cdot \cos(8 \cdot \phi) \\ r^8 \cdot \sin(8 \cdot \phi) \end{bmatrix}
\]

(16)

\[
Z_{xy}(x, y) = Z_{\text{reg}}(\sqrt{x^2 + y^2}, \arctan \left( \frac{y}{x} \right)).
\]

(17)
The difference between designated and measured center of intensity

\[ \Delta M = \begin{bmatrix} C_I(1) - \bar{C}(1) \\ \vdots \\ C_I(q) - \bar{C}(q) \end{bmatrix} \]  

(18)

where \( q \) is the total number of samples, \( C_I(n) \) the measured center of intensity and \( \bar{C}(n) \) the designated center of intensity correlates to the gradient of the function

\[ Z = \begin{bmatrix} \nabla Z_{xy}(x_{cA}(1), y_{cA}(1)) \\ \vdots \\ \nabla Z_{xy}(x_{cA}(q), y_{cA}(q)) \end{bmatrix} \]  

(19)

at the aperture position \( x_{cA}, y_{cA} \). \( \Delta M \) and \( Z \) are then used to calculate the coefficient matrix

\[ K = (Z^T \cdot Z)^{-1} \cdot Z^T \cdot \Delta M \]  

(20)

from which the correction matrix \( P_K \) is calculated. The scaling factor for \( P_K \) depends on the used magnification (X20 objective in first, X40 in second stage) and physical dimension of LCD and CMOS pixels. We determined this scaling factor with a iterative test sequence algorithm in which the minimum of \( \sum |\Delta M| \) is found.

### 3. RESULTS

By applying the previously described analysis methods it was possible to determine wavefront distortions in the optical system and accurate phase shift of a conventional LCD. Figure 7a shows the correction function \( P_K \) after the second stage, which is superposed with the Fresnel lens hologram. The described methods have also been applied to compensate wavefront distortions in a holographic microscope reconstructing the object wave from a diatom and a T-90 Ultra High Resolution Target.

Figure 8 shows the effect to the holographically generated focus. This figure shows the optical imaging of the single-mode optical fiber at the CMOS camera, before and after correction, magnified by a X40 objective. It can be observed how the beam waist on the right side image is considerable smaller.

### 4. CONCLUSION

EASLM even low cost TN-LCDs can be used to create an arbitrary wavefront and simultaneously determine and minimize aberrations introduced by optical components. Different methods for analyzing conventional low-cost
Figure 8. Left side: holographically generated focus $f = 1.5 \text{ m}$. Right side: same focus with phase correction matrix $P_K$ superposed. Pictures are taken with a X40 microscope objective and inverted so dark areas represents high intensity. Camera gain and exposure time on the right image had to be decreased to avoid saturation.

LCD have been presented. It is important to mention that the described method is not limited to transmissive TN-LCDs but can also be applied to other EASLM for example from the LCoS type since the generated wavefront can also be influenced by aberrations.

It has to be shown that the described correction function increases trapping forces in holographic optical traps and the possibility to compensate aberrations could avoid the need for expensive lenses and SLMs. The use of conventional low-cost LCD and standard objectives for holographic optical tweezers can reduce significantly the price of such a device.

Another step is to evaluate and quantify the influence of the described wavefront correction method on image quality like contrast, lateral and depth resolution in DHM with EASLM.

We also need to mention that the radiator fan in the used NEC MT1030G+ beamer introduces vibrations in the whole setup which leads to problems. There were some efforts to decouple them but we will look further to use another TN-LCD for example L3P06S-41G with passively cooled controller board in future projects.

5. ACKNOWLEDGMENTS

This work was supported by ZAFH Photon\textsuperscript{n*}, an association of six universities of applied science and two institutes, which main objective is the further development of photonic processes. ZAFH Photon\textsuperscript{n} is funded by the European Union and the state of Baden-Württemberg.

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