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Interferometric imaging of reflective micro-objects in the presence of strong aberrations

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Abstract: Some imaging techniques reduce the effect of optical aberrations either by detecting and actively compensating for them or by utilizing interferometry. A microscope based on a Mach-Zehnder interferometer has been recently introduced to allow obtaining sharp images of light-transmitting objects in the presence of strong aberrations. However, the method is not capable of imaging microstructures on opaque substrates. In this work, we use a Michelson interferometer to demonstrate imaging of reflecting and back-scattering objects on any substrate with micrometer-scale resolution. The system is remarkably insensitive to both deterministic and random aberrations that can completely destroy the object’s intensity image.

1. Introduction

Reduction of the influence of optical aberrations on image quality has been achieved in multiple imaging techniques, such as adaptive imaging optics [1–3], full-field optical coherence tomography (FFOCT) [4–7], optical scanning [8], and classical ghost imaging (GI) [9–13]. Adaptive optics uses dynamic correction elements, such as deformable mirrors or spatial light modulators to compensate for the specimen-induced aberrations [1]. However, in the case of strong and complex non-deterministic aberrations, this technique may be inefficient. FFOCT is designed for three-dimensional imaging. It uses a Michelson interferometer in which the object replaces a mirror in one of the arms, and the imaging is formed by the interference pattern [14]. The system utilizes a spatially-incoherent light source with a broad spectrum and a short longitudinal coherence length that determines the longitudinal resolution of the system [15]. Therefore, aberrations, if present, should not shift the wave fronts of light by more than this resolution, typically by not more than 10 µm [4,5,15]. Classical GI also uses spatially incoherent illumination. It is, however, based on intensity correlations between two detection arms of the device [16]. The incoherent illumination is usually obtained by passing laser light through a rotating ground glass. Therefore, the longitudinal coherence length can be long, allowing the system to overcome the effects of aberrations stronger than those encountered in FFOCT [17]. However, conventional ghost-imaging setups are complex and have relatively low resolution [18]. Intensity correlations and interference of spatially incoherent light are also used in other techniques, such as single-pixel imaging [19,20] and lensless phase-reversal imaging [21,22] that are able to image both amplitude and phase objects. In particular, image transmission through scattering media has been successfully demonstrated with a single-pixel imaging setup [23,24]. However, a micrometer-scale resolution in the presence of strong aberrations has not yet been demonstrated in these studies.
A novel imaging technique able to retrieve sharp images of transmissive microscopic objects placed behind a strongly aberrating optical diffuser has been recently proposed and demonstrated in practice [25]. The system utilized a Mach-Zehnder interferometer. In the present work, we employ a similar method, but with a different setup that results in a novel universal system for imaging all types of microscopic reflecting and back-scattering objects in the presence of strong aberrations (both deterministic and random) with a micrometer resolution. The system makes use of a Michelson interferometer, and in this sense, it is similar to FFOCT systems. However, the longitudinal coherence length of light in our setup is made an order of magnitude longer than the transverse one. This makes the aberration-insensitive operation of the system similar to that of GI, but based on field interference rather than intensity correlations. We demonstrate theoretically and experimentally that the interferometric microscope is able to show detailed images of both amplitude and phase objects even when the object is screened by an optical diffuser. The technique may therefore be used for imaging micro-objects inside inhomogeneous medical or biological specimens. The key difference of the system from the one based on a Mach-Zehnder interferometer is that it can image not only transparent samples as the previous setup, but also reflecting and back-scattering objects, such as microstructures on an opaque substrate or surface variations of a non-transparent object. Furthermore, in the new system, the input and output parts of the sample arm coincide, which allows one to study surfaces of large objects, including the living ones. In the system with a Mach-Zehnder interferometer, the sample beam passed through the sample that had to be thin and at least partially transparent.

2. Experimental setup

Schematic of the experimental setup is shown in Fig. 1. A light-emitting diode, LED, with an emitting area of $1 \times 1 \text{ mm}^2$ produces unpolarized and spatially highly incoherent light with a peak wavelength of 632 nm and a bandwidth of 18 nm (M625L3, Thorlabs). Light from the source is collimated by a microscope objective $MO_1$ and transmitted through the linear polarizer $P_1$, whose axis can be adjusted to any angle. After propagation through the polarizer, vertical and horizontal components of the electric field vector become mutually correlated and able to form an interference image. A polarizing beam splitter, BS, divides the beam into the reference and sample arms. In the reference arm, a quarter-wave plate, QWP, transforms vertically polarized light into circularly polarized and then into horizontally polarized light on the way back to the beam splitter. As a result, the beam passes through the beam splitter to the camera C (acA 1920-25uc, Basler). The microscope objective with 4× magnification, $MO_3$, which creates the LED surface image in the plane of mirror M, is mounted on a 1D translation stage for tuning the wavefront curvature of the reference beam. Mirror M is also placed on the same stage. The mirror can be moved independently along the optical axis for changing the arm’s length. In the sample arm, an equivalent pair of a quarter-wave plate and a 4× microscope objective, $MO_2$, is used, but the mirror is substituted with a reflecting or back-scattering object, O, to be imaged. The beams from the two arms are combined by the beam splitter and transmitted through a linear polarizer, $P_2$, whose orientation regulates the intensity contributions of the arms to the final image. The combined beam is focused by a lens, L, with a 100 mm focal length to the detector plane of the camera. The system is therefore a polarization-sensitive Michelson interferometer used with a spatially incoherent light source. On the way to the camera, the longitudinal coherence length of light is increased to approximately 0.4 mm by using a bandpass filter, BPF, of 1 nm bandwidth. The filter may also be placed in front of the light source.

In the experiments, optical aberrations are introduced either by shifting the camera from the image plane (creating a focusing error) or by screening the sample with a random static optical diffuser, D. The diffuser simulates a rough surface or a scattering upper layer of a highly aberrating specimen found in, e.g., biology or medicine. When using the diffuser in the sample arm, we inserted a compensating glass plate, CP, of a thickness approximately equal to that of the
Fig. 1. The experimental setup: MO₁, MO₂ and MO₃ – 10×, 4× and 4× microscope objectives, respectively; P₁ and P₂ – linear polarizers, BS – polarizing beam splitter; QWP – quarter-wave plate; M – mirror; O – object; D – diffuser; CP – compensating glass plate; BPF – bandpass filter; L – lens; C - camera. In the reference arm, mirror M and microscope objective MO₃ can be translated along the direction shown by the arrows.

diffuser in the reference arm. The imaging process consists of scanning the reference arm length and recording a short video of the moving interference fringes. Further computer processing of the video allows us to retrieve the object image by calculating the difference between the maximum and minimum signal values at each pixel, which removes the time-independent intensity distribution. More detailed explanation of the object’s image retrieval process is given below. The resolution of the system in the absence of aberrations is determined by the point-spread function of the sample arm. In the described setup, the width of the point-spread function is 0.6λ/NA ≈ 3.8 μm, where the numerical aperture of the microscope objective in front of the object is NA = 0.10.

3. Analysis of image formation

To demonstrate theoretically how the method works we consider the system in terms of scalar wave optics. Let \( R(u, v) \) be the geometric-optics image of the object’s reflection coefficient in the plane \((u, v)\) of the camera detector, \( U(u, v) \) the complex aberration-free image of the field illuminating the sample, which equals the corresponding field in front of the mirror in the reference arm, \( g(u,v) \) the transverse coherence function of \( U(u, v) \), \( h_{im}(u,v) \) the point-spread function (PSF) for light propagation from the object to the camera, and \( \phi_{rs} \) a tunable phase difference of the fields in the two arms of the interferometer before any aberrations are introduced. In the image plane, the fields from the reference and sample arms are

\[
U_r(u,v) = U(u,v)e^{i\phi_{rs}}, \quad \text{and} \quad U_s(u,v) = h_{im}(u,v) \ast [R(u,v)U_{obj}(u,v)],
\]
respectively, where \( * \) denotes convolution. The function \( U_{obj}(u, v) \) describes the field in front of the object to be observed. Its coherence properties can be affected by an aberrative layer covering the object. If no aberration element is present, \( U_{obj}(u, v) \) equals \( U(u, v) \). The width of the coherence function \( g(u, v) \) is approximately given by \( \delta = \delta_0 + 1.2 \Delta z/W_{\text{LED}} \), where \( \delta_0 = 0.6 \lambda / NA \) appears due to the diffraction-limited imaging of the spatially incoherent source (equal to the width of the point-spread function of the system without aberrations), \( \Delta z \) is the distance from the plane of the perfect image of the source along the optical axis, and \( W_{\text{LED}} \) is the size of the LED’s image. The second term in the expression for \( \delta \) is due to gain of coherence by propagation [26]. The width of \( h_{\text{im}}(u, v) \), on the other hand, is \( \delta_{\text{im}} = \delta_0 + 2 \Delta z NA \). Here, \( \delta_0 \) is the same diffraction-limited width of the point-spread function in the focal plane, as above, and the second term is the additional width away from the focal plane.

The intensity of the total field in the image plane is

\[
I(u, v) = \langle |U_s(u, v)|^2 \rangle + \langle |U_c(u, v)|^2 \rangle + 2 \text{Re}\{\langle U_s(u, v)U_c^*(u, v) \rangle\},
\]

where the angular brackets stand for time averaging. The interference term, in which \( U(u, v) \) and \( U_{obj}(u, v) \) are the only time-dependent random functions, can be written as

\[
\langle U_s(u, v)U_c^*(u, v) \rangle = \int_{-\infty}^{\infty} h_{\text{im}}(u, v) e^{-i \phi} dxdy.
\]

In the case of a focusing error (obtained, for example, by shifting the camera), the functions \( U(u, v) \) and \( U_{obj}(u, v) \) are proportional to the same random process. Therefore, we can write

\[
\langle U_{obj}(x, y)U^*(x, y) \rangle = \delta_{\text{im}}(u, v) g(u, v) R(x, y) \langle U_{obj}(x, y)U^*(x, y) \rangle e^{-i \phi_0}. \]

Here we used the fact that a LED field can be described by the Schell model [27]. Hence, we obtain

\[
\langle U_s(u, v)U_c^*(u, v) \rangle = I_1 e^{-i \phi_0} \int_{-\infty}^{\infty} h_{\text{im}}(u, v) g(u, v) R(x, y) dxdy,
\]

where \( \phi_0 \) is independent of the transverse coordinates but depends on the arm-length difference.

The interferometric image is formed by the difference between the maximum and minimum values of \( I(u, v) \), when the length of the reference arm is scanned. The image is given by

\[
I_{\text{im}}(u, v) = 4I_1 \int_{-\infty}^{\infty} h_{\text{im}}(u, v) g(u, v) R(x, y) dxdy,
\]

or

\[
I_{\text{im}}(u, v) \propto |h_{\text{im}}(u, v) g(u, v) * R(u, v)|.
\]

For a severe focusing error, \( h_{\text{im}}(u, v) \) is wide compared to \( g(u, v) \). For example, if \( \Delta z = 1 \) mm, the width of \( g(u, v) \) is \( \delta = 5 \) mm, while the width of \( h_{\text{im}}(u, v) \) is \( \delta_{\text{im}} = 204 \) \( \mu \)m (we recall that \( \lambda = 632 \) nm, \( NA = 0.1 \) and \( W_{\text{LED}} = 1 \) mm). The width of \( h_{\text{im}}(u, v) g(u, v) \) will therefore be close to \( \delta = 5 \) \( \mu \)m and the interferometric image will be sharp. The intensity image of any microscopic object, on the other hand, will be washed out, because the imaging resolution is equal to \( \delta_{\text{im}} = 204 \) \( \mu \)m.

If the object is covered by a random aberrating layer (such as a diffuser), the function \( U_{obj}(u, v) \) differs from \( U(u, v) \) by a wider and more complex coherence function. This function is determined by the point-spread function \( h_{\text{obj}}(u, v) \) that describes propagation of light through the diffuser to the object. On average, the width of \( h_{\text{obj}}(u, v) \) can approximately be written as

\[
\delta_{\text{obj}} = \delta_0 + 2d \sin \theta,
\]

where \( d \) is the distance between the object and the diffuser and \( \theta \) is the diffusing angle of the diffuser [28]. This approximate equation is obtained by considering the diffuser as an array of microlenses with the numerical apertures equal to \( \sin \theta \). The width \( \delta_{\text{obj}} \) can be much larger than \( \delta \). In this case, we can assume that \( U(u, v) \) is delta-correlated and that the field distorted by the diffuser is \( U_{obj}(x, y) \approx U_{obj}(x, y) * U(x, y) \). This allows us to write
\[
\langle U_{\text{obj}}(u,v)U^*(u,v) \rangle \approx h_{\text{obj}}(x,y) \ast \langle U(x,y)U^*(u,v) \rangle = I_1 h_{\text{obj}}(x,y) \ast \delta(u-x,v-y) = I_1 h_{\text{obj}}(u-x,v-y).
\]
Hence, as follows from Eq. (4) the retrieved image intensity is given also by Eq. (7), but with \( g(u,v) \) being replaced with \( h_{\text{obj}}(u,v) \). The image resolution will be lower, because \( h_{\text{obj}}(u,v) \) is wider than \( g(u,v) \). However, we still have an improvement of the resolution, as long as \( |h_{\text{obj}}(u,v)| \) is narrower than \( |h_{\text{im}}(u,v)| \). Note that the resolution will be higher for a diffuser with smaller \( \theta \) positioned closer to the object.

When imaging a transparent phase object with a reflection coefficient \( R(u,v) = e^{i\phi_R(u,v)} \), the function \( I_{\text{int}}(u,v) \) drops at the object edges. Let us assume for simplicity that the object has a straight edge that coincides with the \( v \)-axis in the image plane and that the phase shift \( \phi_R(u,v) \) is equal to 0 for \( u<0 \) and \( \pi \) for \( u>0 \). Then, the reflection coefficient can be written as \( R(u,v) = 1 - 2H(u) \), where \( H(u) \) is the Heaviside step function. Similarly to the previous case, Eq. (7) yields
\[
I_{\text{int}}(u,v) \propto |h_{\text{im}}(u,v)h_{\text{obj}}(u,v) \ast [1 - 2H(u)]|,
\]
where \( h_{\text{obj}}(u,v) = g(u,v) \), if the aberrations are weak or caused by a deterministic focusing error. Because \( 1 - 2H(u) \) switches its value from 1 to -1 when crossing the point \( u = 0 \) and the function \( h_{\text{im}}(u,v)h_{\text{obj}}(u,v) \) is localized, the image of the edge is a dark line along the \( v \)-axis. The intensity decreases when \( u \) approaches 0. The resolution, or the width of the line, is for a focusing error determined by the width of \( g(u,v) \). If the object is located behind a random scattering film, \( h_{\text{obj}}(u,v) \) becomes wide compared to \( g(u,v) \) and speckled, and the image intensity gains random variations. The object can still be recognized through the dark contours described by Eq. (8).

The influence of aberrations on the image formation can be summarized as follows. In the imaging system, the used transversely incoherent illumination field in front of the sample is identical to that in front of the reference mirror. The interference fringes in the image plane can be created only by the waves originating from the same points of the two illumination fields. Due to the sample-arm aberrations, each of these points imaged via the sample arm is blurred. This increases the static background intensity component of the image and decreases the fringe modulation depth. However, the computer processing removes the static background component, so that only the decreased fringe modulation amplitude is observed in the final image. Aberrations can also make the fringe contrast inhomogeneous. In some areas of the image, they can longitudinally shift the sample-arm wavefronts by a distance larger than the longitudinal coherence length, in which case the fringe pattern disappears. In view of this, to accommodate for larger aberrations, one should use a source of longer temporal coherence. In addition, interference fringes disappear, if aberrations shift the imaged sample points transversely by a distance larger than the transverse coherence length. To compensate for this effect, one can increase the transverse coherence length by shifting the source along the optical axis, which will, however, worsen the resolution. If the aberrations spread the images of each point considerably, the mutually correlated fields overlap in the image plane even in the presence of the mentioned transverse shift.

4. Experimental results

Three types of samples were fabricated to test the proposed method and setup: reflecting, transmitting and back-scattering. Two reflecting amplitude-modulated samples represent the logo of the University of Eastern Finland. They were made by patterning an Al film on glass with a laser writer (LW405, MICROTECH). The fabricated amplitude-modulated samples are the negatives of each other. The logo in Fig. 2(a) is formed by metal stripes on glass and the one in Fig. 3(a) is made of slits in a metal film. The width of the metal stripes is 12 \( \mu \)m, while the width of the slits is 20 \( \mu \)m. The third sample is a transparent phase-modulated sample that was fabricated by patterning a 1.5 \( \mu \)m thick layer of photoresist on glass. It represents the logo of Aalto University formed by 13 \( \mu \)m wide photoresist stripes (see Figs. 2(d) and 3(e)).
Fig. 2. Optical images of the amplitude-modulated logo of the University of Eastern Finland (a) and the phase-modulated logo of Aalto University (d). The images are obtained without aberrations and with the reference arm blocked (a) and open (d). Pictures (b) and (e) show the object image obtained from the sample arm with induced focusing error. Images (c) and (f) are the ghost-like interferometric images retrieved from the interference pattern of the reference beam and the disturbed object beam.

Fig. 3. Optical images of an amplitude-modulated logo (a) and a phase-modulated logo (e) obtained without aberrations. Pictures (b) and (f) show the images obtained from the sample arm after covering the sample with the optical diffuser. The third and the fourth columns contain the interferometric images of the samples retrieved from the interference patterns. The diffuser is placed in contact with the object in the third column, and at a distance of a few millimeters in the fourth column.

A severe focusing error makes the intensity images formed by the sample arm disappear, as shown in the middle column of Fig. 2. We start by setting the interferometer’s arm lengths equal. The focusing error was obtained by shifting the camera along the optical axis back from the image plane by a few millimeters. Alternatively, the sample could be shifted, in which case, however, the arm-length difference of the interferometer would change. Next we open the reference arm and adjust the position of the microscope objective to sharpen the reference point-spread function on the camera. The arm-length difference stays unchanged. Recording and processing the interferometric data, we obtain images shown in the right column of Fig. 2. The images are almost as sharp as the original, aberration-free images. They contain all the details of the amplitude-modulated logo and reveal the sharp edges of the phase-modulated logo, as the theory in the above section predicts.
Next we test imaging of a slit sample (Fig. 3(a)) and a phase-modulated sample (Fig. 3(e)) in the presence of strong random aberrations caused by a static optical diffuser. The second column in Fig. 3 shows the effect of the diffuser on the intensity image obtained from the sample arm. Using the reference arm and the image retrieval procedure, we then obtain the images shown in the third column. Even though the diffuser makes the background of the objects highly uneven, the objects are considerably sharpened in the retrieved images. If the object is unknown, it can be difficult to differentiate between the structures of the object and the background pattern caused by the diffuser. However, if the object structures are not random, as in our case, they can readily be resolved. The background can be made smoother by shifting the diffuser a few millimeters away from the sample, as demonstrated by Fig. 3(d) and Fig. 3(h). This, however, also decreases the image resolution.

In order to test imaging of a back-scattering object with a curved surface, we used a sample in the form of a thin metal wire (80 µm in diameter) placed on a tape (see Fig. 4(a)). An optical image of the sample illuminated with the LED light without aberrations can be seen in Fig. 4(b). Because the wire has a cylindrical shape, it looks bright at the centre and dark on the sides. A large focusing error essentially destroys the image, as seen in Fig. 4(c). However, opening the reference arm and making use of the image retrieval, we successfully obtain the aberration-free image shown in Fig. 4(d). This experiment demonstrates the ability of the system to perform aberration-insensitive imaging of curved surfaces of three-dimensional objects as well.

To show the ability of the system to work also in the FFOCT regime, we conducted an experiment on imaging a part of an electronic chip with approximately 50 µm surface height variation. The intensity image of the object is shown in Fig. 5(a). To increase the resolution we replaced the system’s microscope objectives with the ones having a higher numerical aperture of 0.25 (δ₀ ≈ 1.5 µm). The left-hand side of the object is about 50 µm farther away from the objective than the right-hand side, and the dark vertical stripe is a tilted surface between the two horizontal surfaces. We sharpen the image area close to the left edge of the tilted surface, leaving the other areas blurred. Applying the interferometric image retrieval technique, we observe the object with both the bottom and top surfaces imaged with a high resolution in spite of the focusing error (see Fig. 5(b)). Then we remove the band-pass filter and switch the system to the FFOCT regime. This reduces the longitudinal coherence length of light to about 10 µm. As a result, we can shift the sample along the optical axis and obtain sharp interferometric images of 10 µm thick slices of the object one by one. Figures 5(c) and 5(d) show sharp images of the bottom and the top surfaces of the structure, respectively. However, they cannot be observed simultaneously in this regime. This example illustrates the advantage of our approach over the classical FFOCT.
Fig. 5. Imaging a two-level microchip surface. (a) The ordinary intensity image obtained with the reference arm blocked. Here the right-hand side is elevated above the left-hand side by about 50 µm. The dark vertical stripe in the middle is a tilted surface between the two levels. The image is sharp only along the left edge of the tilted surface. (b) The retrieved interferometric image. The resolution is high for both the bottom and top surfaces. In (c) and (d), the images are obtained without the band-pass filter at two different reference-arm lengths, with the length difference of 50 µm.

5. Conclusion

To conclude, we introduced a method for imaging all types of reflecting and back-scattering micro-objects in the presence of strong aberrations. The method also successfully works with highly transmissive phase-modulated and reflective three-dimensional objects. Compared to our previous interferometric system based on the Mach-Zehnder interferometer, the one proposed in this paper is capable of imaging microstructures on an opaque substrate. Furthermore, since the input and output parts of the sample arm coincide, surfaces of large objects can also be imaged. The imaging principle can be applied, e.g., in endoscopy. We demonstrated both theoretically and experimentally that the arrangement is remarkably insensitive to deterministic and random optical aberrations and can be switched to the FFOCT regime to obtain additional information on the surface topology. In particular, the system can be used to obtain sharp images of surfaces with large height variations. We foresee applications of the presented imaging approach in microscopy dealing with micro-objects inside scattering and aberrating samples, such as biological and medical specimens.

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Disclosures

The authors declare no conflicts of interest.

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