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Controlling light in scattering media using ultrasound modulation

Blanca Mestre Torà

Supervised by Dr./Prof. Martí Duocastella, (UB)

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Blanca Mestre Torà
Advisor: Martí Duocastella
Departament de Física Aplicada, Facultat de Física, Universitat de Barcelona, Diagonal 645, 08028 Barcelona, Spain.
E-mail: blanca.mestre@estudiantat.upc.edu

Abstract.
Light-based methods are fundamental in biology and biomedical sciences. They allow for non-invasive diagnosis and localized treatments with resolutions down to the sub-cellular level. However, light scattering inside biological tissue constrains the maximum depth at which current methods can operate. In this work, we show that light can be focused and controlled in scattering media using ultrasound waves. We experimentally prove how ultrasound waves can work as embedded lenses in the scattering media, helping to compensate for scattering and redirect light toward a deeper focus. Our results demonstrate qualitatively and quantitatively how such ultrasound-enabled lenses enable up a factor of 7 improvement in light focusing for samples with a scattering coefficient of 3.5 mm^{-1}, compared to traditional focusing with external elements. This allows to resolve images of test samples immersed in scattering media - USAF target with spacing of 27.8 μm - that would be completely hidden with existing methods.

Keywords— ultrasound, scattering, optical microscopy, laser surgery

1. Introduction

The scattering of light is a ubiquitous phenomenon in nature. Produced by the local variations in refractive index distributions inside a material, it prevents the focusing of light deep inside heterogeneous media such as biological tissue [1, 2]. As a result, current light-based methods are only optimized to operate at depths below a fraction of a millimeter inside tissue [2]. In more detail, when light propagates inside tissue, the number of ballistic photons - photons not deviated - decreases exponentially until they become negligible after a certain propagation distance called the transport mean free path (TMFP) [3], see figure 1a. Such a distance depends on the wavelength of light and material properties. Even when operating in the so-called biological windows - located at near-infrared spectral region [4] -, the TMPF for skin is only 1.1 mm and for lungs, it is even lower (0.6 mm) [2]. Deeper focusing and control of light is needed to obtain information of internal sections inside tissue. Given the relevance of light-based methods as diagnosis and treatment tools in biomedical and life sciences [5, 6], it is imperative to develop new methods for light focusing in scattering media.

Over the recent years, several techniques have been developed to address the light scattering issue, including wavefront shaping (WFS) methods and endoscopy. The core idea of WFS is that scattering is a deterministic process. Thus, if we know information about the tissue, we can undo scattering by modulating the incident wavefront so that scattered photons constructively interfere at a target position [7]. There are three types of WFS depending on the employed method to calculate the optimal input wavefront: feedback-based wavefront shaping [8], transmission matrix inversion [9], and optical phase conjugation (OPC)/optical time reversal [10, 11]. For these techniques, accessing the inner specimen sections is always required, independently of the method used to determine the optimal incoming wavefront [7]. In practice, however, this is a difficult task, especially when minimally invasive approaches are desired. They also require time to retrieve information about the scattering tissue. Another method for deep light focusing in tissue is endoscopy. It consists of using fiber bundles - called endoscopes - that are directly inserted into the body. While allowing for an unlimited penetration depth, they are intrinsically invasive. Therefore, deep light focusing with existing methods always comes at the expense of increased invasiveness [12], loss of spatial and/or loss of temporal resolution [3].

In this work, we propose a novel approach for focusing and guiding light into scattering media with reduced invasiveness and high spatiotemporal resolution. The proposed technique is based on using ultrasound waves to periodically modulate the optical properties of a medium. More precisely, we use a resonant cylindrical cavity to spatially and temporally modulate the refractive index of such medium. This modulation effectively acts as an embedded waveguide or lens inside the medium, helping to compensate for scattering and redirecting light towards a deeper focus. By using a customized imaging system, such as a camera or a photodiode with virtual lock-in detection, we characterize the effect of ultrasound modulation on light focusing, and quantify the improved imaging resolution - both in transmission and reflection modes - for different samples and scattering coefficients.

2. Light focusing with ultrasound

The core concept of our approach is to induce a nonuniform refractive index distribution in the scattering medium by using ultrasound. Ultrasound are pressure waves created by the compression and rarefaction of a medium. Such changes in pressure instigate local variations of the medium density, and consequently, of its refractive index [13]. This phenomenon, known as the acousto-optic effect, has been used to generate variable focusing elements by filling a cylindrical piezoelectric cavity with an homogeneous liquid [14]. Our hypothesis is that, by filling the cavity with a scattering medium, such ultrasound focusing will compensate for the deviation of photons, acting as an embedded waveguide or lens that helps to provide deep focusing, as shown in figure 1b.

In this project we selected a cylindrical cavity with an inner diameter of 16 mm and a length of 20 mm made of the piezoelectric material PZT (lead zirconate titanate). When filled with a liquid and driven with a radio-frequency that matches with one of the radial resonant frequencies of the cavity (in the range of 3-5 MHz), stationary Bessel modes appear inside the cavity [15]. The induced refractive index changes in the liquid are given by (1).

$$n(r,t) = n_o + n_A \cdot J_0(kr) \cdot cos(\omega t + \phi), \tag{1}$$

where n_o corresponds to the static refractive index of the liquid, n_A to the amplitude of the refractive index change - it depends on the driving amplitude voltage -, J_0 is the Bessel

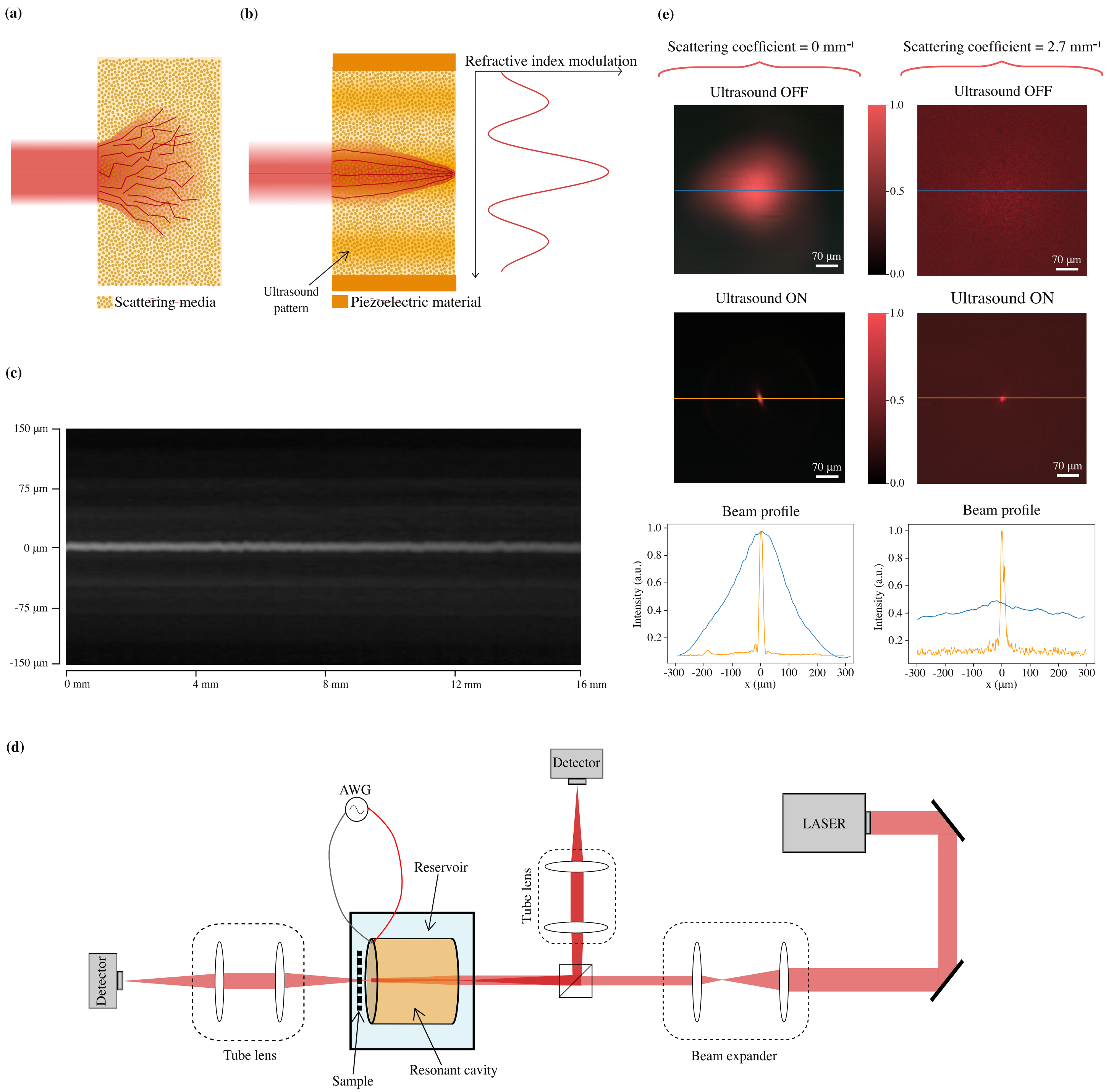

Figure 1: Acoustic focusing of light. (a) Schematic of the scattering process that light undergoes when incident in scattering media. (b) Schematic of the proposed method for light controlling in scattering media. (c) Image of a longitudinal cut of the beam propagating after the resonant cavity. (d) Image of the experimental set up used in this project. (e) Images and profiles of the beam at the output of the resonant cylindrical cavity.

function of first kind, k is the acoustic wavevector, and ω to the driving frequency. The overall effect of such refractive index upon incident light is to induce a phase change, in analogy to a variable gradient index of refraction (GRIN) lens [16]. An incident Gaussian beam traversing this refractive index is then focused into a Bessel-like beam [17], as shown in figure 1c. A Bessel-beam is characterized by a central lobe that can propagate long distances without spreading - above 16 mm in our experiments. They also exhibit self-healing properties, making them more robust against scattering [18]. Notably, the temporal dependence of the refractive index inside our cavity (1) can produce, at a given time instance, either a Bessel-like beam focused along the optical axis or along a ring. To operate always with the former, synchronized pulsed light is necessary. Alternatively, an average effect is obtained with continuous illumination, which results in less light intensity along the optical axis.

A scheme of the customized system that we implemented to focus light with an ultrasound cavity is shown in figure 1d. A diode laser with a wavelength of 660 nm (Coherent Obis) is employed as the light source, which beam is reduced with a 4f system of 0.2X magnification. The beam is guided to the piezoelectric cylindrical cavity using various optical elements. To enable rapid selection of the medium filling the cavity, we designed a transparent chamber where we could place the cavity with micrometric precision. After the cavity, we placed the sample and sample holder. The later is attached to an xyz stage with a precision of 0.2 μm (Pysick Instrumente) controlled with a LabVIEW program. The system can be operated with either transmitted or reflected light. The transmitted beam is directed toward a 150 mm tube lens, after which we placed a light-sensitive detector - a CCD camera, or an avalanche photodiode (APD), both from Thorlabs. The reflected beam is redirected via a beam splitter toward a second tube lens (150 mm) and a second light-sensitive detector - a second CCD camera, that enabled imaging both reflected and transmitted light simultaneously, or the APD. The data extracted from either sensor is processed in a custom LabVIEW program, which allows displaying images of the sample in real-time.

An example of the light-focusing effects with ultrasound is shown in figure 1e. All images correspond to the beam at the output of the cavity acquired with the CCD. The upper ones correspond to the cases when the ultrasound is off for the cavity filled with water (left), and with a water/milk solution (right). The striking difference between the two images is due to the effect that scattering has in light propagation. Indeed, water is a homogeneous liquid and thus the beam can propagate with no significant distortion - the measured full width at half maximum (FWHM) of the beam profile at its center is about 300 μm, the size of the beam at the input of the cavity. Instead, the water/milk mixture is highly scattering* - by analysis of the light attenuation as a function of thickness [19], the scattering coefficient value in this case was 2.7 mm^{-1} (right). The result of light scattering produces an attenuation of the light intensity and a spreading of the beam spot. Importantly, the beam spot size is a key parameter in optical microscopy, since it determines the resolution of an optical system - the spot size is directly linked to the so-called point spread function (PSF). The observed deterioration in light confinement inside the water/milk mixture proves the negative effects that scattering can have in light focusing and imaging applications.

Bellow (figure 1e), the images correspond to the focused beam via the embedded lens in the media when the ultrasound is on for the cavity filled with water (left), and with a water/milk solution (right). Compared to the previous case, where the ultrasound in the

* The water/milk mixture enables easy tuning of the scattering coefficient by simply changing the concentration of milk in the solution, and is thus selected as the model for the scattering medium in all experiments hereby.

cavity was off, now, in both cases the focused laser beam can be discerned from the background and the beam diameter - at the output of the focusing element - is close to 30 μm, which is maintained in the cavity filled with a scattering media of 2.7 mm^{-1}. The maintained shape of the focused beam inside water/milk solution is due to the ultrasound waves redirecting scattered light to a deep focus.

3. Evaluation of optical properties

First, we evaluated the focusing performance of the resonant cavity operating as an embedded lens/waveguide in a medium. To this end, we measured the modulation transfer function (MTF). The MTF is a parameter that can be used to measure the ability of an imaging system and its capability of reproducing fine detail. The MTF values indicate the contrast of captured objects as a function of the spatial frequency [20]. Usually, the MTF decreases when increasing the spatial frequency, down to a value where objects are not discernible - the so-called cutoff frequency, which determines the maximum spatial frequency the optical system can resolve, namely, the resolution of the system [20, 21]. Typically, the cut-off frequency is defined at an MTF value of 0.1 [21, 22].

We characterized the MTF by using the slanted-edge method. As its name indicates, this method is based on capturing images of a knife-edge target [22], which are processed following three steps, shown in figure 2a. First, a line profile from the sharp edge region of the image is retrieved, generating the edge spread function (ESF). From the ESF, the line spread function (LSF) of the system is calculated by taking the numerical derivative of the ESF sample. Finally, the MTF is calculated from the normalized Fourier Transform of the LSF [23].

In this work, we study the optical properties of our focusing system for several scattering media mixtures. Initially, we operate the laser source in pulsed mode. The light pulses are synchronized with the stationary modulation of the refractive index inside the cavity, so that light only propagates when the ultrasound cavity is working as a focusing element. In this case, we use a CCD camera as a detector.

Figure 2b (•) shows the spatial resolution of the system (MTF cut-off frequency) operated in transmission mode as a function of the scattering coefficient of the water/milk mixture when focusing with ultrasound (piezoelectric cavity driven at 4 MHz and an amplitude voltage of 20 V_{pp}) along the medium. Remarkably, the spatial resolution of the system is around 25 μm and it remains constant as the scattering coefficient of the medium is increased. To better contextualize these results, we compared them with the classical way of focusing light, that is, using an external optical element. For a fair comparison of both methods, as the external focusing element we use the same ultrasound cavity but filled with water. Thus, the Bessel-like beam is generated before entering the scattering medium. As shown in figure 2b (•), in this case the spatial resolution rapidly deteriorates with the scattering coefficient. More precisely, the spatial resolution of the system goes from nearly 25 μm (the same value as with ultrasound focusing along the medium) to 170 μm as the scattering coefficient is 0 mm^{-1} and 3.5 mm^{-1} respectively. Therefore, for inhomogeneous media, the use of ultrasound focusing can lead to an enormous gain in spatial resolution - up to a factor of 7 in current experiments.

While the ultrasound cavity within the inhomogeneous medium seems to not be affected by scattering, we noticed that the number of background photons significantly increases with the scattering coefficient. Therefore, the lens embedded inside the scattering media may maintain its focusing capability, but the number of ballistic photons decreases with the scattering coefficient. Such an effect will ultimately limit the imaging capability of the system -

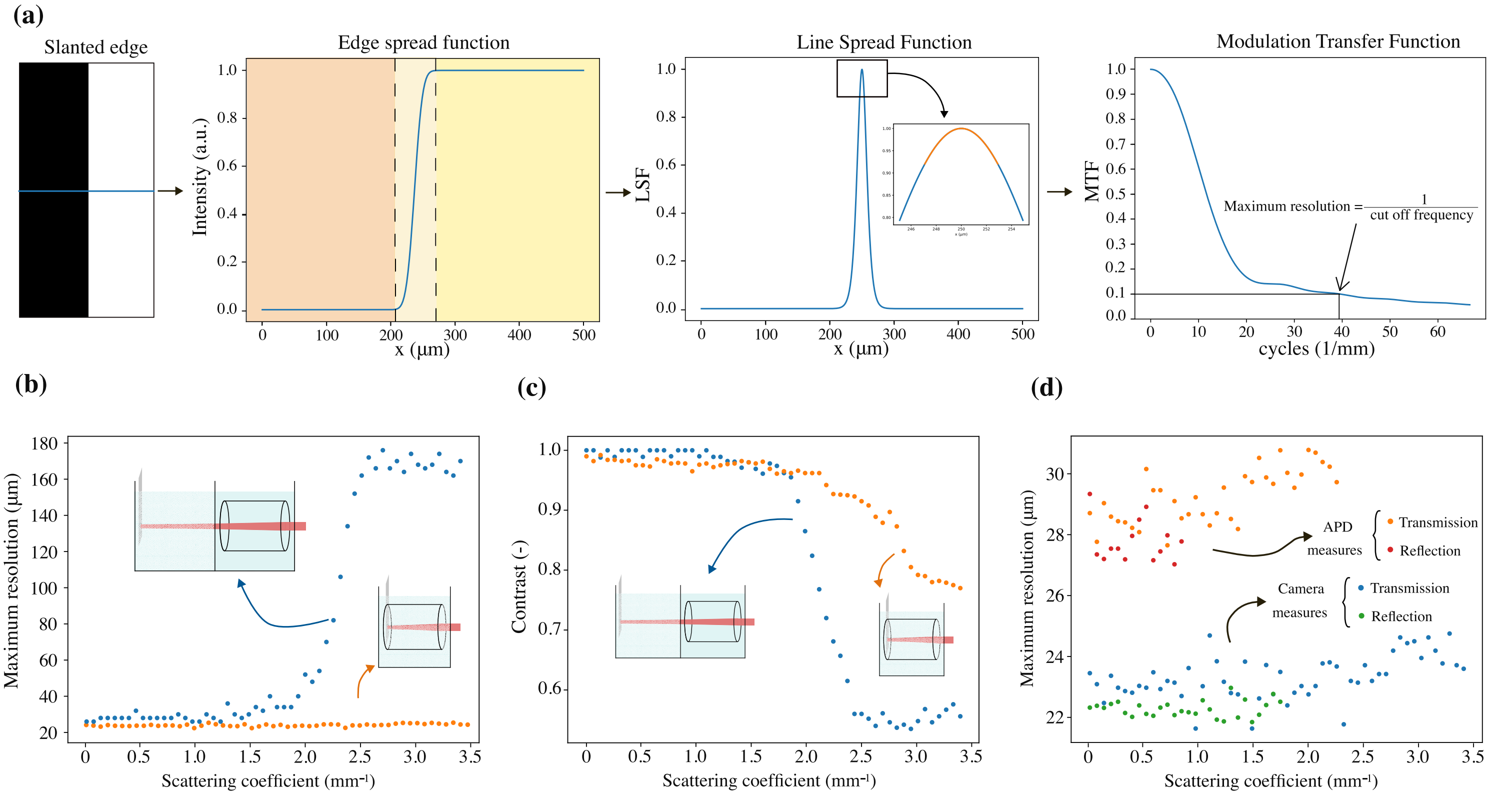

Figure 2: Optical performance of the ultrasonic cavity for light focusing. (a) Scheme of the process to measure the modulation transfer function (MTF) of the optical system via the slanted edge method. (b) Optical resolution and (c) contrast of the focusing system working as an internal (●) or external (●) lens at different scattering conditions. (d) Optical resolution of the embedded focusing system when working in reflection and transmission mode, whether using a CCD camera or an APD as a detector.

eventually, the ratio of ballistic vs background photons would be too low to discern an object. To quantify this phenomenon, we introduce a contrast parameter defined as the difference in light intensity between the ballistic photons (I_{max}) and the adjacent background (I_{min}) relative to the overall intensity ($I_{max} + I_{min}$) [22], see (2).

$$Contrast = \frac{I_{max} - I_{min}}{I_{max} + I_{min}} \tag{2}$$

As expected, the contrast function extracted from the slanted-edge images decreases with the scattering coefficient (figure 2c). Still, there is a clear benefit of using ultrasound focusing. When using an external element for focusing (figure 2c (●)), the contrast decreases rapidly, from nearly 1 to 0.5 as the scattering coefficient of the mixtures varies from 0 mm^{-1} to 3.5 mm^{-1} respectively. Instead, during the same interval, the contrast function when using ultrasound focusing only decreases about 20% (figure 2c (●)).

The previous results show promise for ultrasound focusing. However, in most imaging applications involving biological tissue constructs, access to transmitted light is not possible. To this end, we characterized the system response when operating in reflection mode. In this case, the light is being focused as long as it travels through the scattering media until it reaches the sample at the end of the cavity. The reflected light propagates in the media until leaving the chamber - note that, in this configuration, the light traverses the scattering medium twice. Following the same protocol discussed before, the evaluation of the optical performance is determined by the slanted-edge method. Figure 2d (•) shows the system resolution as a function of the scattering coefficient. As in the previous case, the resolution of the system when using ultrasound focusing is maintained constant at a value of 25 μm. Interestingly, though, measurements were only possible up to a value of the scattering coefficient of 1.8 mm^{-1}, approximately a factor of 2 smaller than in transmission (•). This is expected given the longer length the light has to travel inside the scattering medium, and hence the faster deterioration of the signal to background ratio.

Finally, we decided to explore the effects of ultrasound focusing with continuous light. Such operation greatly simplifies implementation given the possibility to obviate for the complex synchronization between ultrasound and pulsed light. In this case, we used the APD for measurements. Thanks to the high-speed of the APD, we can capture the modulation frequencies induced by the ultrasound (4 MHz), obtaining a readout signal that is sinusoidal. Note that only the ballistic photons along the optical axis will be strongly modulated. Thus, using a custom virtual lock-in amplifier implemented in LabVIEW, we can separate these photons from the scattered (and not modulated) photons. As shown in figure 2d (•) for transmission and figure 2d (•) for reflection, the spatial resolution, measured from the MTF, is around 29 μm in both cases. Such a value remains approximately constant with the scattering coefficient, in agreement with previous results obtained using synchronized pulsed light. The reported differences in spatial resolution (25 μm) vs 29 μm can be explained by the "average" effect of the generated Bessel-like beam when using CW light - not only less light is focused along the optical axis, which can help to explain the decrease in the maximum scattering coefficient value that can be used, but the effective focused spot size increases.

4. Proof of concept

To check the possibility of using ultrasound focusing for imaging, we acquired images of a resolution target (1951 USAF test chart) immersed in different scattering media. We selected the chart regions corresponding to the group 3 element 5 - with elements consisting of bars with a width of 39.37 μm -, and the group 4 element 2 - with a bar width of 27.84 μm - close to the spatial resolution of the system that is 25 μm. All images are acquired using point-by-point scanning. In more detail, the samples under observation are scanned around the focused illuminating beam, which is in pulsed mode and synchronized with the focusing cavity. In this case, we use a CCD camera as a detector. The extracted data from the latter is processed and the images are reconstructed in a custom LabVIEW program in real-time.

Figure 3a and figure 4a show images of the resolution target when immersed in pure water. The left images are acquired with the focusing system working as an embedded lens/waveguide, and the right ones using an external focusing element. As expected, when using homogeneous media there are not significant differences between images acquired with either method. In contrast, the results are stunningly different when using scattering media, as shown in figures 3b, 3c, 3d, and 4b. When using ultrasound focusing, we can distinguish in all cases the bars with approximately the same resolution, albeit with a loss of contrast. When using the external focusing element, instead, there is a significant deterioration of resolution as the scattering coefficient of the medium increases. This can be noted in the right images of figures 3b, 3c, 3d and 4b - for a scattering coefficient of 3.3 mm^{-1} the lines are not resolvable at all. These results experimentally demonstrate the possibility of ultrasound focusing for imaging in scattering media, validating the initial hypothesis.

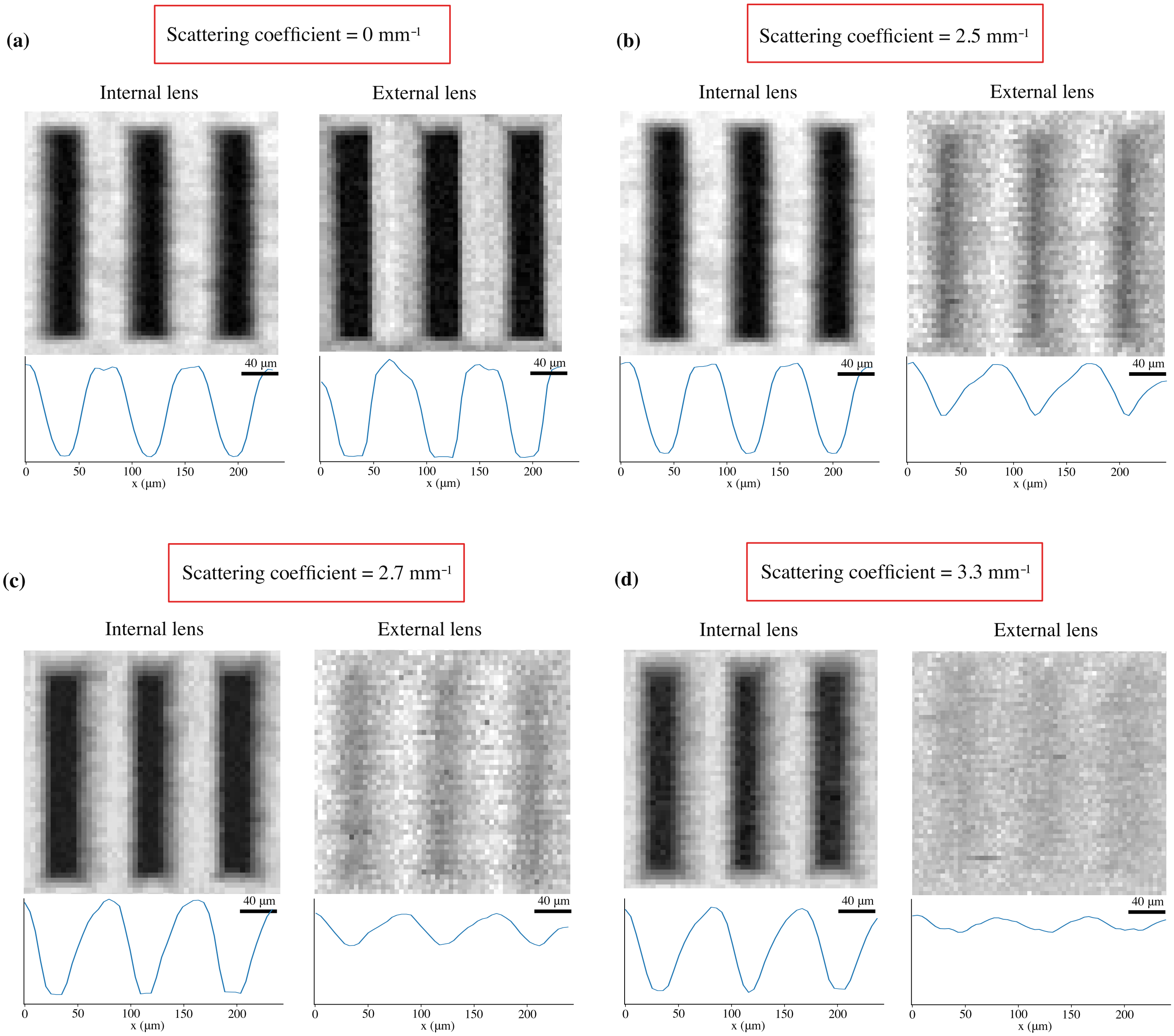

Figure 3: Experimental validation of the ultrasound focusing of light. The images refer to the USAF target group 3 element 5. Each set corresponds to different scattering conditions in the medium. The pixel size of the images is 5 μm. The average profile of the bars is shown bellow each image as well.

5. Conclusion and future work

Controlling light with ultrasound waves in inhomogeneous media enables overcoming the scattering of light. As our results demonstrate, ultrasound waves can be used to focus light down to 25 μm in scattering media with a scattering coefficient of up to 3.5 mm^{-1} - better resolutions are expected by using more intense ultrasound. Instead, when focusing the light from an external source, the spatial resolution can be up to 7 times worse, with an abrupt decrease in contrast. Compared to existing methods, ultrasound focusing offers a high spatio-

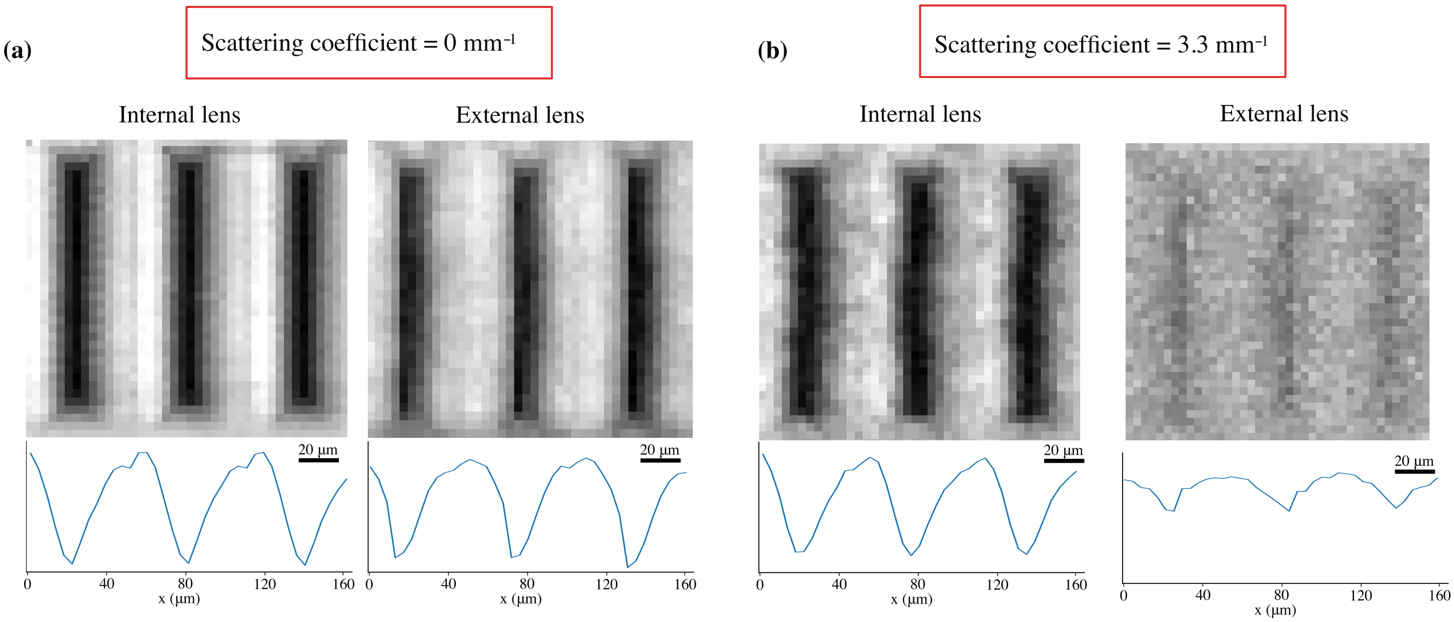

Figure 4: Experimental validation of the ultrasound focusing of light. The images refer to the USAF target group 4 element 2. Each set corresponds to different scattering conditions in the medium. The pixel size of the images is 4 μm. The average profile of the bars is shown bellow each image as well.

temporal resolution and, provided pressures below 10 MPa are used, it can be regarded as a safe an non-invasive technology.

In future work, it would be interesting to develop a less restrictive method for producing refractive index modulation inside scattering media. Instead of working with a resonant cylindrical cavity, which produces stationary sound waves only inside of it, a source producing travelling sound waves and further modulation of the refractive index could be considered.

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