



Matrix Approaches in Acousto-Optic Imaging

Louis Dutheil

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DE L'UNIVERSITÉ PSL

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IMAGING is a tool at the core of medical diagnosis, as it provides key elements to understand physiological and metabolic phenomena. As early detection has a crucial importance for the success of many disease treatments, current research on imaging is committed to overcome the resolution limits of existing techniques. These improvements walk together with the increase of the sensitivity, that provides a clearer information to physicians to build their diagnosis. The portability of medical imaging systems also participates to early detection. As an example, the development of even more compact ultrasound scanners has improved their implementation in medical practices or emergency vehicles, bringing it closer to the patients. Work to reduce the invasiveness diminishes the risks of drawbacks for the patient and also the duration of imaging. The diversity of imaging techniques brings complementary information as many viewpoints on the medical case.

In that context, optical imaging techniques has many advantages. They are non-invasive — endoscopes use optical fiber to introduce inside the body with practically no impact to probe the surface of organs — and high resolution — microscopes have sub-micrometer resolution — allowing to monitor internal activity in cells. A large variety of optical techniques can be harnessed to probe biological phenomena and thus understanding metabolic functions. Indeed, as chromophores, inner biological markers such as hemoglobin, absorb light at different wavelengths, we can directly observe metabolic functions by measuring the absorbance of tissues. Thus, optics allows vascularization and functional imaging in organs. The light propagation through biological tissues is also characterized by multiple scattering. It means that after a millimeter of propagation in tissues, light loses the memory of its initial direction and polarization. This phenomenon imposes a limit to standard optical imaging such as microscopy or coherent tomography in most of human tissues. Ophthalmologic imaging is an exception to this phenomenon as it benefits from the limpid window of cornea. Providing a direct access to inner tissue, ophthalmologic examination can thus inform on micro-vascular diseases. Although, after a millimeter, the optical information is no longer directly accessible.

Multi-wave imaging brings solution to this optical limit imposed by scattering. Relying on the coupling of ultrasonic and optical waves in soft tissues, photoacoustic tomography and acousto-optic imaging benefit from the both advantages of ultrasound scanning and optical contrast. Indeed, ultrasounds have a ballistic propagation in tissues which allows remote emission or reception without a loss of the acoustic resolution. Moreover, the optical contrast gives access to biological and metabolic information. Acousto-optic imaging thus uses ultrasound to guide the optical imaging beyond a millimeter in depth. This method consists in focusing an ultrasonic wave within a sample illuminated by a laser source. Where the acoustic and the optical waves overlap, part of the photons are spectrally shifted by the frequency of the ultrasounds. As a result, everything appends as if there were virtual source of known location buried inside the medium. As the amplitude of this virtual source is proportional to the local absorption, the tagged-photon detection gives access to the optical contrast in the waves interaction area. Scanning the ultrasound focus allows mapping the local optical properties of the sample, such as absorption or scattering. Therefore, acousto-optic imaging gives access to an optical information deep into scattering media with an acoustic resolution.

The main difficulty of acousto-optic imaging concerns the detection. Indeed, the tagged-photons contribution is extremely close in frequency to the untagged ones, and thus almost impossible to resolve spectrally with conventional spectrometers. Different techniques exist to filter the tagged-light contribution. In this work, we will use digital holography, the scattered light is being collected by a camera sensor. Using a local oscillator at the tagged-photon frequency, we get rid of the main untagged-light contribution. Therefore, as an interferometric method, holography remains on coherent light illumination. This method relies on common technologies, camera detection, which makes it quite light to implement. Nevertheless, the coherence dependency of this technique makes it sensitive to temporal and spatial decorrelation occurring in biological media. It means that, because of the stochastic motion of structures at multiple scales, the interferogram impinging the sensor is totally renewed after a characteristic time of 1 ms. In order to image *in vivo*, the tagged-light detection needs to be performed within a millisecond range. For this, we implement a basis of acoustic excitations providing enough tagged signal to be robust to temporal decorrelation.

Moreover, digital holography gives also access to the phase of the spatial wavefront of the tagged photons. Then, using a spatial light modulator, we can monitor the spatial wavefront at the input of the medium. The scattering sample can be seen as a linear process linking input and output optical components, the acousto-optic interaction acting as a spatial constraint on the propagation. Therefore, we apply the transmission matrix theory on it to identify best transmissive eigen-channels. This thesis gives matricial approaches in acousto-optic imaging with a first viewpoint on the acoustic excitation and a second on the acousto-optic transmissive channels through the optical system.

This work is organized in four chapters. The chapter one sets the scene of light propagation in biological media. We present different optical and hybrid imaging techniques; diffuse optical tomography and photoacoustic tomography. After introducing the mechanisms of the acousto-optic effect, we demonstrate how holographic camera-based

detection allows selecting the tagged light.

The second chapter focuses on the ultrasonic excitation. We develop different temporal modulations used in imaging with spatially-focused ultrasounds. Then, we move to emitter arrays and spatially-structured ultrasounds, heading to matricial approach of the excitation.

The third chapter heads to the acousto-optic transmission matrix. After introducing the general formalism of transmission matrix of optical systems, we explain how it is a tool for focusing into scattering media through time-reversal. We finish by describing the singularities of the acousto-optic transmission matrix.

The final chapter of the thesis presents the development of the experiment on which we measure the acousto-optic transmission matrix. We explain how the principal component analysis allows us to focus inside the acoustic focal area beyond the theoretical limit imposed by the acoustic diffraction limit.

CHAPTER 1

Optical imaging in multiple scattering media

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MEDICAL imaging is a key tool for the diagnosis provided by physicians and specialists. A large diversity of techniques take advantage of different ranges of the electromagnetic spectrum. These techniques provide information on the optical contrast within the tissues. This contrast reflects inhomogeneities of the medium that are linked to differences of properties depending on the physical phenomena brought out by the imaging modalities. To that extent, optical contrast carries a structural information of the area crossed by the light.

In this first part, we present the properties of light propagation in biological tissues. We see how acousto-optic imaging allows overcoming the barrier of multiple scattering regime to probe the optical contrast in depth within the medium. After presenting the acousto-optic effect, we detail the holography technique to measure the amplitude and the phase of an optical field using a reference beam as a local oscillator. Then, we explain how holography allows detecting the optical field of virtual light sources created under the acousto-optic effect.

1.1 Optics in biological media

In this first section, we give an overview of the main optical phenomena taking place in biological tissues.

1.1.1 Optical therapeutic window

The first constraint, that optical imaging in biological media faces, is the absorption property of tissue. According to the Beer-Lambert equation:

$$I(z) = I_0 e^{-\mu_a z}, \quad (1.1)$$

the intensity of light I decreases exponentially with depth z within the tissue. This decrease is characterized by the absorption coefficient μ_a which depends on the wavelength λ of the optical wave. This absorption coefficient varies for the different optically-active constitutive elements of biological tissue. These elements, called chromophores, have parts of their molecules that capture light energy. Water and hemoglobin are the two predominant chromophores in human tissues, their spectral-absorption profiles are presented on [Figure 1.1](#). These profiles clearly show a gap between 600 and 1300 nm which corresponds to the range of wavelength where the light has its maximum depth of penetration in biological tissues. This interval of low absorption is called the optical therapeutic window. Near-infrared light is thus privileged to observe in these tissues.

1.1.2 Scattering

Another phenomenon affecting optical imaging in biological tissues is scattering. This phenomenon is due to the presence of optical index inhomogeneities of variable sizes within the medium. As a consequence of these complex variation of index, the paths followed by the light in its propagation are very difficult to predict based on the

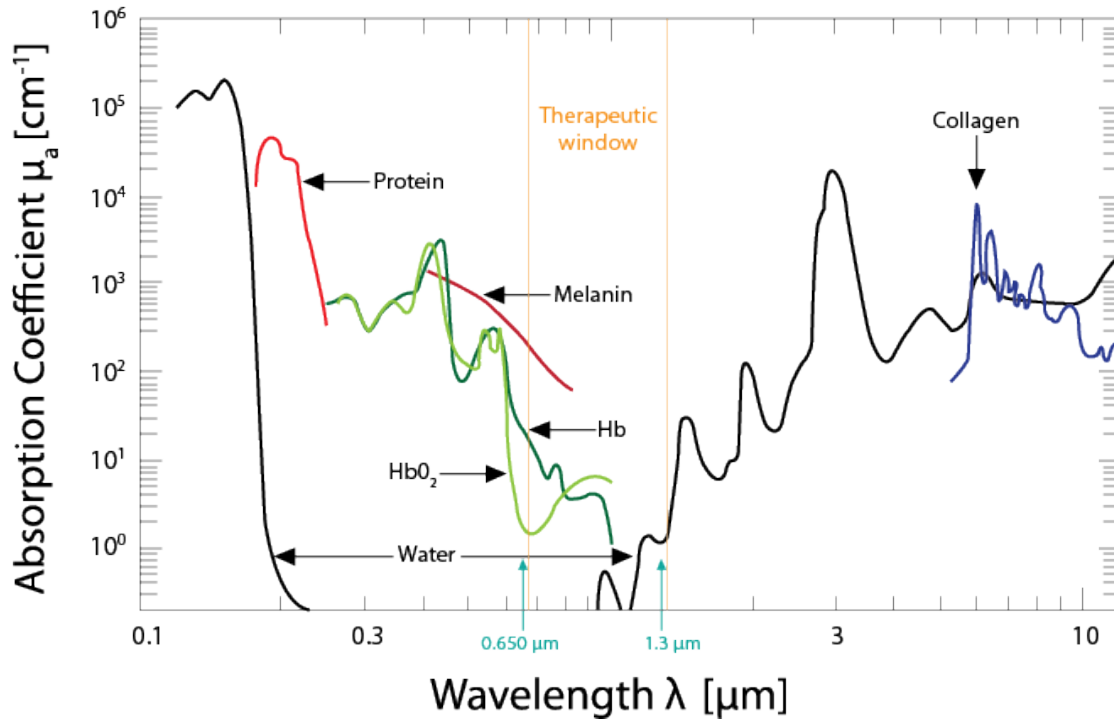


Figure 1.1 – Optical therapeutic window in biological tissues (LiteCure, 2018). Hb and HbO₂ stand respectively for the hemoglobin and the oxyhemoglobin, which are physiologically dissociable combination of hemoglobin with dioxygen, playing the role of carrier in blood vessels and giving them their red color.

direction of the incident beam. The dominant propagation regime switches from ballistic to scattering and multiple scattering. As a result, the localization of an optical information is no longer accessible. Optical multiple scattering theory is explained in many works such as [Ishimaru \(1978\)](#), [Akkermans and Montambaux \(2007\)](#). Two main theories explain it: the Rayleigh theory, for scattering particles smaller than the wavelength of the optical wave, and the Mie theory, for larger particles. These theories are incomplete in order to describe light propagation in biological media as they combine a size diversity of structures.

Nevertheless, multiple scattering can be understood from a mesoscopic point of view as a distribution of scatterers, i.e. scattering events homogeneously distributed across the medium. The distribution properties are qualified by the scattering mean-free-path, denoted l_s , which corresponds to the characteristic distance traveled by light between two consecutive scattering events. The inverse of the scattering mean free path, namely the scattering coefficient, $\mu_s = l_s^{-1}$, given in cm^{-1} , characterizes the scattering at the macroscopic level, $l_s = 100 \mu\text{m}$ for biological tissues.

1.1.3 Scattering anisotropy

A second properties of light propagation in biological media is anisotropy. Indeed, because of the typical size of the scatterers, larger than the wavelength of the optical wave, the scattering of light follows Mie's theory. Thus, light is predominantly scattered forward at each scattering event. The statistical direction of deviation is quantified by the anisotropy factor g , defined as the average cosine of the deviation angle θ with respect to the direction of the incoming photon (see [Figure 1.2](#)).

$$g = \langle \cos(\theta) \rangle. \quad (1.2)$$

As we have said, scattering is mainly forward and the anisotropy factor is typically between 0.7 and 0.9. In this condition, light does not loose its polarization and its global direction as it undergoes a scattering event. We need to define another coefficient, the transport mean free path l^* , taking in account both the spatial recurrence of scatterers and their anisotropy:

$$l^* = \frac{l_s}{1 - g}. \quad (1.3)$$

This quantity characterizes the distance after which light has lost the memory of its original direction and polarization. In biological tissues, the transport mean free path is equal to 1 mm. Its inverse, the reduced scattering coefficient $\mu'_s = l^{*-1}$, expressed in cm^{-1} , macroscopically characterizes the anisotropic scattering. The [Equation 1.3](#) emphasizes the role played by the anisotropy in the multiple scattering regime. A wider anisotropy, characterized by $g \approx 0$ can change the order of magnitude of the transport mean free path.

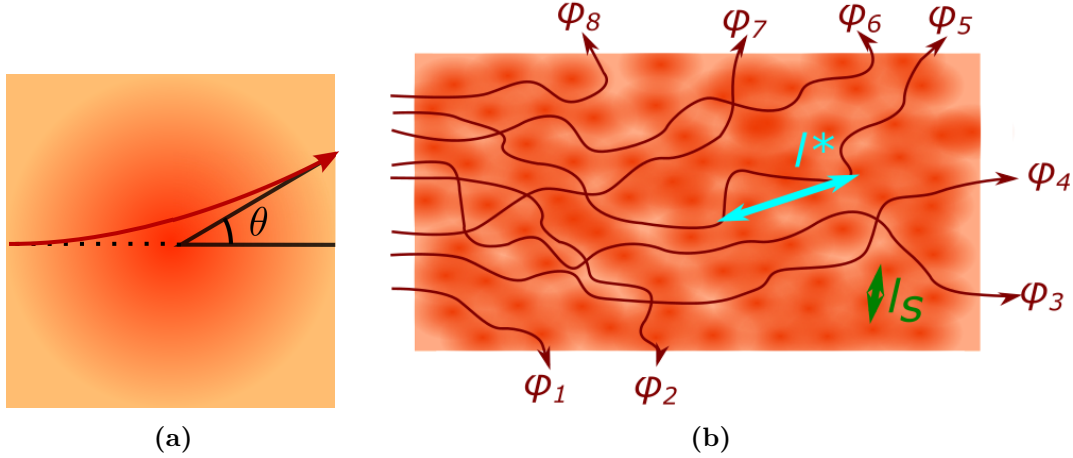


Figure 1.2 – (a) Schematic of a scattering event. Statistically, depending on the shape and the power of the scatterer, light is deviated by an angle θ when it crosses the scatterer area. The scatterer is the manifestation of optical index inhomogeneities inside the medium, depicted by a color gradient on these schematic, such as changes of composition, density, pressure. (b) Schematic of a multiple scattering medium in which scatterers are randomly distributed, separated each other by the typical distance l_s , the scattering mean free path. Considering a collimated beam incoming to this medium, the light paths are highly distorted by scatterers. As a result, the direction of light is totally lost beyond the typical depth l^* , namely the scattering mean free path of transport.

1.1.4 Propagation regimes

The characteristic lengths given previously have to be compared with the thickness of the sample in order to estimate the predominant propagation regime. For this, we distinguish three propagating regimes based on the ratio between the thickness W of the medium and the scattering mean free path of transport l^* :

- for $W \ll l^*$ the light barely interacts with scatterers as it passes through the medium. The propagation is dominated by the ballistic regime, the optical information is localized. This range corresponds to the validity domain of standard microscopy techniques and full-field optical coherence tomography, i.e. few hundreds of micrometers. These imaging techniques have a wavelength resolution lower than or equal to the micrometer.
- the $W \sim l^*$ domain corresponds to the simple-scattering regime. Supposing the Born approximation satisfied, the index variation between the medium and the scatterers is weak with respect to 1. The beam spreads in a cone. Photons are multiply scattered but along snake-like paths. The initial direction of propagation is partially kept.
- the $W \gg l^*$ domain corresponds to the multiple-scattering regime. All the pho-

tons undergo multiple scattering. This case corresponds to biological media beyond 1 mm in depth.

Biological tissues present a large range of structures at different levels. From the nanometers of cyto-skeleton rods to the few micrometers of cells and group of cells. This diversity of multiscale structuration is fruitful for optical scattering phenomenon. Moreover the structure of tissues varies across the body, which makes different values for the quantities presented earlier. In the literature, multiple studies have probed the optical properties of multiple tissues. They have been gathered by some reviews (Cheong et al., 1990; Jacques, 2013). Table 1.1 gives the amplitude range of the optical properties. Then, in some tissues, the optical properties are affected by the development

n	Refraction index	1.35 to 1.45
μ_a	Absorption coefficient	1 cm^{-1} to 10 cm^{-1} at 630 nm
μ_s	Scattering coefficient	100 cm^{-1} to 500 cm^{-1} in the visible domain
g	Anisotropy factor	0.7 to 0.9
μ'_s	Reduced scattering coefficient	2 cm^{-1} to 70 cm^{-1} in the visible domain

Table 1.1 – Optical properties in human tissues found in the review of Cheong et al. (1990). These data have been measured at 630 nm, wavelength closed to the maximum penetration depth range (see Figure 1.1).

of diseases. As these anomalies are partially transparent for other imaging techniques, optical imaging appears as a key technique to diagnose these pathologies. As an example, Grosenick et al. (2005) detail the optical properties of 87 carcinomas, combined with mammographic detections. They revealed that absorption is twice higher in carcinomas, probably due to the higher vascularity in these tissues, and that the reduced scattering coefficient is globally 20 % higher than the surrounding healthy tissues.

Hence, optical imaging provides added values compared to other techniques used for medical diagnosis. Indeed, as it is directly sensitive to optical changes in the tissues it has a great specificity and unveils anomalies at an early stage which is a crucial advantage for prevention and the success of treatments. In the next sections, we present how we retrieve local information from the scattered light.

1.2 Speckle

At the output plane of a multiple-scattering medium shone by a monochromatic plane wave, the optical beam is widely spread. As a result of the multiple light paths inside the medium, the phase in the output plane is highly heterogeneous. Out coming from the medium, this complex pattern undergoes free propagation. Then, if we consider a plane parallel to the output surface of the medium located at a distance L , the sum of phasors — i.e. complex wavelets with different phases and incidences accumulated along different optical paths — gives rise to a complex figure of interference that we call speckle. This interference pattern consists of a succession of bright and dark spots resulting from the constructive and destructive sum of these wavelets (see Figure 1.3).

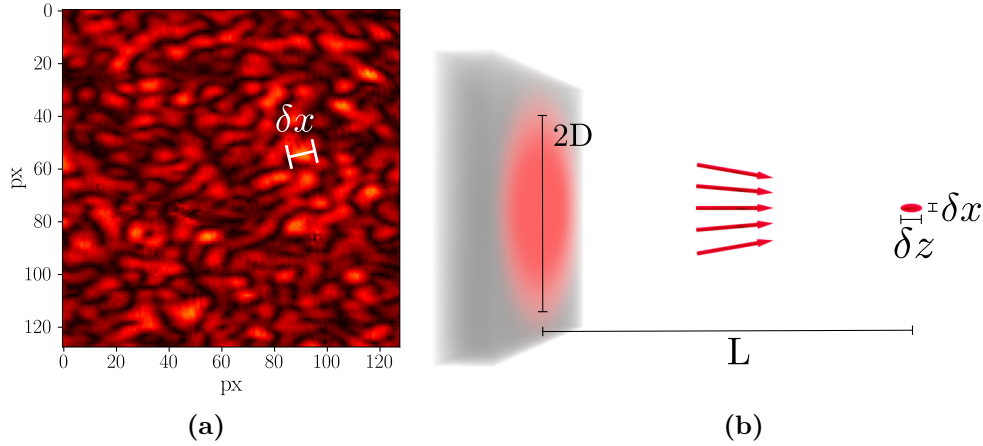


Figure 1.3 – (a) Speckle pattern recorded by the camera. (b) Speckle grain size from a given distance L at the output of a scattering medium. The numerical aperture in the far-field, i.e. $D/L \ll 1$ is $\text{NA} = D/L$

Bright spots are called speckle grains. Their typical size depends both on the wavelength of the monochromatic wave, and the numerical aperture NA of the scattered spot on the output-plane of the scattering medium seen from the plane of observation.

$$\text{Transverse resolution} \quad \delta x \sim \frac{\lambda}{\text{NA}}, \quad (1.4)$$

$$\text{Axial resolution} \quad \delta z \sim \frac{\lambda}{\text{NA}^2}. \quad (1.5)$$

The speckle pattern presented on the left side of [Figure 1.3](#), has been recorded on a camera placed at 60 cm from the output surface of a highly-scattering hydrogel illuminated by a 780 nm-monochromatic laser source. The output optical field is spatially filtered by a 6 mm radius diaphragm. As a numerical application of [Equation 1.4](#), we can calculate the typical size of a speckle grain:

$$\delta x = \frac{0.780 \times 600}{6} = 78 \mu\text{m}. \quad (1.6)$$

This roughly corresponds to 6 pixels of the camera (pixel pitch $\Delta_{\text{pix}} = 13.6 \mu\text{m}$).

The speckle carries out the phase of the optical paths in its interference pattern. Many works have focused on the statistical properties of the speckle to retrieve patterns hidden behind a scattering layer. As an example, [Katz et al. \(2014\)](#) exploit the correlation of the speckle to retrieve the image of an object hidden behind a highly-scattering medium. This work is based on the memory effect of the scattering medium. This effect corresponds to the preservation of the speckle pattern produced by an object hidden behind a scattering medium, for small displacement of the object. This means that two light rays coming, by a slightly-opened angular range to the scattering medium, undergo the same optical path through it before leaving with a slightly-different angle. Hence, the interference figure in the far-field contains multiple partial information on the object

transmitted through the medium by all the scattering channels. Auto-correlating the speckle pattern, and thanks to a phase-retrieval algorithm, the authors reconstruct the image of the object. In an other study, [Batarseh et al. \(2018\)](#) harness spatial coherence of a speckle produced by reflection on a white-painted wall with no *a priori* knowledge on the light source.

In further sections, we see how to retrieve the phase and the magnitude of the optical field from this complex figure based on holographic techniques.

1.3 Optical imaging techniques

We now present optical imaging techniques in multiple scattering media. We begin with diffuse optical tomography.

1.3.1 Diffuse optical tomography (DOT)

Diffuse optical tomography (DOT) is an all-optic method based on the detection of diffuse light. The input illumination is provided by an array of light sources, and detection is handled using an array of detectors placed on the surface of the imaged object ([Välisuo, 2015](#)). The light sources used are in the near-infrared to match the therapeutic window.

The measurement is performed by recording the light flux collected on each detector by successively illuminating the sample by each light source. As relying on the measure of the light flux, DOT is robust to speckle decorrelation. Processing the different signals allows retrieving information on the optical propagation. For this, the propagation volume is meshed into unit elements. The recorded signals are taken as boundary conditions to solve the inverse problem in order to infer localized tissue parameters ([Durduran et al., 2010](#)). The reconstruction is mainly based on a random walk as the propagation model. Its working hypotheses are a quasi-isotropic radiance, an isotropic source and a small temporal variation of the luminous flux in a rotation symmetry. Moreover, this principal model is corrected close to air/tissue interfaces, very anisotropic or absorbent tissues, such as liver, and low scattering environment. Once retrieved, the map of the optical properties can be linked to physiological parameters such as the absorption by chromophores. As one of the dominant chromophores in biological tissues, hemoglobin and oxyhemoglobin concentrations are key markers to diagnose vascularization and oxygenation of organs or deep skin structures, such as tumors. The performances of DOT are constrained by the spatial sampling of the detector distribution and the accuracy of the reconstruction algorithm. DOT has a millimeter resolution and a maximum observation depth of 6 cm ([Dalla Mora et al., 2015](#)). Moreover, imaging in depth is done at the expense of the signal-to-noise ratio.

Within the two last decades, major improvements allowed compacting the detectors and reducing the global cost of the technique ([Pifferi et al., 2016](#)). This evolution gives rise to portable system and fiber-based sources, these devices can be harnessed to illuminate organs, providing *in situ* imaging. A number of *in vivo* applications and

clinical studies were possible in different areas, such as optical mammography (Grosenick et al., 2016), brain mapping (Torricelli et al., 2014) and muscle monitoring (Contini et al., 2012). Different DOT systems exist using continuous or structured illumination in combination with different photon detections such as temporal or spectral filters (Pifferi et al., 2016; Dalla Mora et al., 2020).

Because the diffuse photons lost their directional information, all-optical techniques have a limited depth of observation in biological tissues. In order to propose an in-depth optical imaging, one needs to take advantage of multiple-wave interaction. As mechanical waves, used in echography, undergo ballistic propagation in biological tissue, in the following section, we present techniques coupling acoustic and optic waves.

1.3.2 Photoacoustic tomography (PAT)

The parenthood of acousto-optic imaging and photoacoustic tomography as multi-wave imaging techniques are often confused with each other by students. Their principle are quite different however. A schematic of the principle of PAT is presented on Figure 1.4. Photoacoustic tomography uses a intense Q-switched nanosecond laser pulse to illuminate the sample. As light propagates through the sample, it undergoes absorption. Under photoacoustic effect, the pulse energy is absorbed by chromophores, which then relax through thermal radiation (Wang and Yao, 2016). The consecutive raise of temperature causes the local dilatation of the medium, creating a mechanical wave in the megahertz range, that propagates through the sample. This acoustic wave is recorded at the surface by multiple transducers or an echographic probe. The acoustic signals are then processed to retrieve the image of the optical-energy distribution absorbed by the tissues. One identifies two main sources of contrast for PAT which are similar to DOT's ones. The first are chromophores naturally present in tissues. We have introduced them in the subsection 1.1.1, namely water, hemoglobin, oxyhemoglobin and deoxyhemoglobin, proteins and collagen (Xia et al., 2014). Hemoglobin has high interest as it quantifies the vascular circulation allowing functional imaging (Na et al., 2021; Hosseinaee et al., 2021). Then, artificial agents such as antibody-conjugated gold-nanoparticles provide a sharp contrast (Abelha et al., 2020). PAT have been studied at the Langevin Institute (Bossy et al., 2006; Gateau et al., 2013; Chaigne et al., 2014; Jayet, 2015). This techniques is active and take advantages of wavefront shaping and matricial approaches for sharpening its resolution these last years (Sun et al., 2019; Poudel et al., 2019; Inzunza-Ibarra et al., 2020).

1.4 Acousto-optic effect

In the previous section, we have seen that despite the multiple scattering effect, light conveys exploitable information through complex media. It is possible to deduce properties of the hidden object by processing the complex speckle pattern produced. We present now how the acousto-optic effect allows probing the contrast within such scattering media.

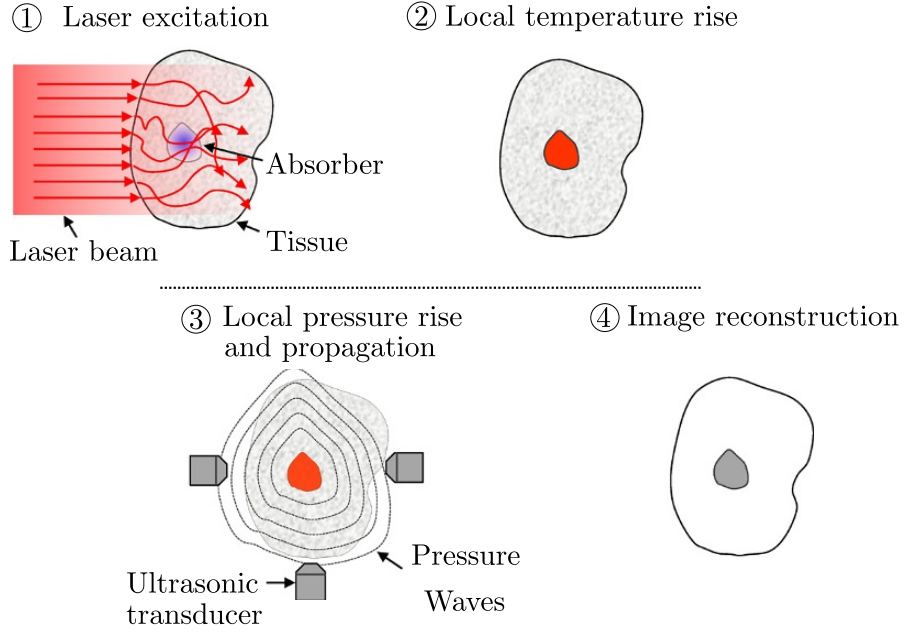


Figure 1.4 – Principle of photoacoustic tomography. The laser excitation is absorbed locally by chromophores leading to thermal radiation. The resulting temperature rise creates a pressure wave that propagates across the sample. The pressure front sampled by a ultrasonic transducers. Spatially processing the recordings allows retrieving the absorption contrast image.

Historically, the effect of elastic waves on the propagation of light in the same media have been predicted by Brillouin (1922). Few years later, Debye and Sears (1932) and Lucas and Biquard (1932) experimentally observed the diffraction of light at it propagates throughout a crystal submitted to acoustic waves. Therefore, the ultrasonic pressure wave propagating in the medium hence induces a diffraction grating by locally modifying the optical index, namely the acousto-optic effect. This effect has two manifestations on the light as depicted on Figure 1.5, an angular deviation and a frequency shift.

Under normal incidence, light is diffracted with the order n at the angle θ_n and at the frequency f_n , as described by Equation 1.7:

$$\begin{aligned}\theta_n &= \pm \arcsin \left(\frac{n \lambda}{\Lambda} \right), \\ f_n &= f_L \pm n f_{US},\end{aligned}\tag{1.7}$$

with Λ the acoustic wavelength, λ the optical wavelength and n the diffraction order. Acousto-optic modulator, used commonly in optical setups as deflectors or optical switches, are based on this diffraction principle. In our work we use them, as we will see later, in order to shift in frequency the optical beams.

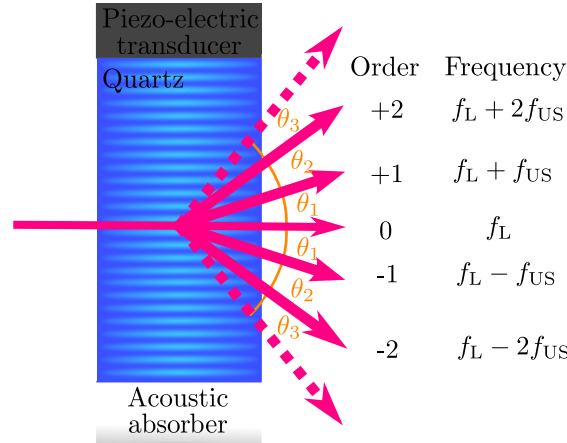


Figure 1.5 – Principle of the acousto-optic effect, a piezo-electric transducer creates an ultrasonic wave in a quartz crystal. The pressure wave locally modifies the optical index and induces a diffraction grating. This dynamic grating diffracts a normal incident monochromatic light into orders. In addition, the dynamic structure induces a Doppler effect which diffracts many orders, each being shifted in frequency. This phase modulation can be seen as a superposition of sinusoidal moving gratings with different frequencies and wave vectors, as the decomposition of a phase modulation.

An additional effect occurs in scattering media. Due to the pressure wave which alternatively compresses and relaxes the medium, scatterers oscillate and modulate the optical paths (Leutz and Maret, 1995; Wang, 2001) (see Figure 1.6). This effect plays at unison with the Doppler's effect induced by the optical index modulations explained above. As a result, at the output of the scattering medium affected by the propagation of ultrasounds, the spectral density of the photons crossing the insonified region is split into two side-bands surrounding the central carrier frequency of the laser (see Figure 1.6). We call the photons shifted in frequency at $f_L \pm f_{US}$ respectively the positively and negatively tagged photons. As the acousto-optic effect is localized where the ultrasound waves are, the amplitude of the detection of tagged photons provides a local information on the optical contrast. Acousto-optic tagging provides sort of virtual controllable sources within the medium of study, whose amplitude is proportional to the local intensity of the scattered light.

1.5 Spectral hole-burning filtering

In contrast to interferometric measurements, spectral hole-burning filtering of the tagged-photons (SHB) is a detection method based on the measurement of a flux. It consists in generating a narrow-band filter able to discriminate ultrasonically-tagged photons over a large etendue. This technique, originally inherited from Solid State Spectroscopy, uses rare-earth doped inorganic crystal at liquid helium temperature. Rare-earth ele-

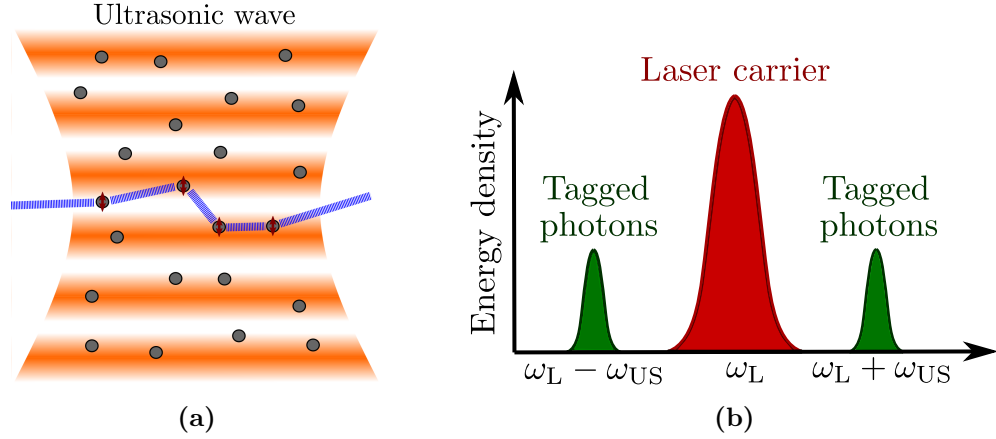


Figure 1.6 – (a) Schematic of the acousto-optic effect in scattering media, the ultrasonic pressure wave propagating in the medium, alternatively compresses and relaxes the medium. This local motion simultaneously affects the position of scatterers and modulate the optical paths. (b) Spectral density profile of photons crossing an insonified region

ments correspond to scandium (Sc), yttrium (Y) and lanthanides (Ln), a family of 14 elements included between lanthanum (La) and lutetium (Lu) whose electronic structure is:

$$[\text{Xe}]6s^24f^n \quad 1 \leq n \leq 14. \quad (1.8)$$

Lanthanides have close chemical properties. In particular, their stable oxidation state is +III in general, which corresponds to the loss of $6s^2$ electrons and one $4f$ electron, giving the final electronic structure $[\text{Xe}]4f^{n-1}$. Thus, the last filled energy levels are in the $4f$ electron shell, more internal than $5s$ and $5p$ ones and thus benefit from shielding. This gives to optical transitions, such as absorption or fluorescence, a high spectral sharpness at low temperature. Then, the homogeneous width denotes the full-width at half-maximum (FWHM) of the absorption line of a single ion. For example the homogeneous width equals 3 kHz at 1.5 K in Tm:YAG (Venet, 2019). Rare-earth doped crystals used in SHB are artificially synthesized by growing the crystal with a controlled composition and doping of the rare-earth. When incorporated in the matrix host, the energy levels of the ions are slightly distributed, depending on the electrostatic environment of the substitution sites (Macfarlane, 2002). As a consequence, the energy levels spread inhomogeneously along a spectral domain called the inhomogeneous linewidth. Depending on the matrix topology, doping ions and concentration, the inhomogeneous linewidth varies between few dozens of megahertz to hundreds of gigahertz. The filter is made by ‘burning’ a spectral hole within the inhomogeneous linewidth of a rare-earth doped crystal by saturating the optical transition with a spectrally narrow laser. This laser beam, called pump, saturates a transition and stores the atoms in a reservoir level, inducing a local absorption modification within the spectrum. This hole in the absorption profile creates a transparency window (see Figure 1.7).

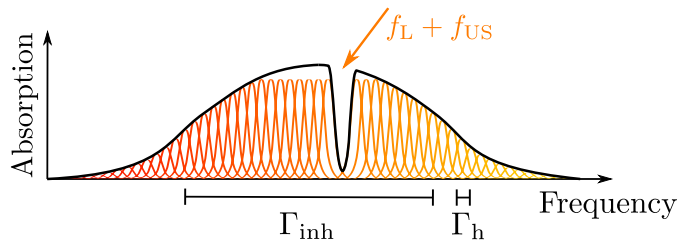


Figure 1.7 – Principle of spectral hole-burning filtering. The hole burnt within the absorption profile at the tagged-photons frequency ($f_L + f_{US}$) favors their transmission towards the detector, while un-shifted photons (f_L , majority) are absorbed by the crystal. Γ_{inh} denotes the inhomogeneous linewidth and Γ_h the homogeneous linewidth.

In order to have sharp homogeneous lines — necessary to correctly filter the pertinent photons as the difference between the carrier and the sidebands is of some megahertz — the crystal needs to be maintained at a cryogenic temperature. As we have seen in the [subsection 1.1.1](#), biological imaging imposes to use optical sources in the therapeutic window. Thus, the SHB transition is required to be in this window. Yttrium aluminum garnet crystals doped with thulium ions (Tm:YAG) have an inhomogeneous absorption line centered on 793 nm. This makes them suited for biological imaging and adapted to laser diodes. They have been used in an acousto-optic imaging context for the first time by [Li et al. \(2008\)](#), providing a 14 dB-attenuation of the laser carrier frequency compared to the tagged-photons. With this system, a metastable level having a lifetime of 10 ms is involved in the burning process. Within the last years, many improvements have been made to stabilize the filter hence extending its lifetime. Using a magnetic field through the crystal at 2 K, [Venet et al. \(2019\)](#) performed persistent SHB, with a hole lifetime longer than a second. This extended lifetime allows ‘burning’ a deeper hole. Moreover, it allows stopping the pump beam during the acquisition, which causes stray light on the detector.

Praseodymium-doped yttrium orthosilicate (Pr:YSO) have been proposed for acousto-optic imaging by [Zhang et al. \(2012\)](#). Pr:YSO belongs to a special class of SHB materials, called persistent SHB. They have long hole lifetime, seconds or longer, which allows the use of complex hole-burning pulse sequences in order to engrave deep spectral holes. These filters provide very good performances with scattering sample, allowing the probe of few-centimeter-thick hydrogels. Indeed, a deep spectral hole also has a steep refractive index dispersion at its center, which gives a slow group-velocity for the tagged light signal. These persistent SHBs are so-called slow-light filters. As a consequence, time-gating the acquisition allows dark-background detection of the tagged light. Nevertheless the inhomogeneous absorption line is centered on 606 nm, wavelength for which no laser diode exists and is located on the edge of the therapeutic window. The swedish team of Stefan Kröll makes continuous improvement in the development of persistent SHB filters. They provide even efficient carrier attenuation, from 30 dB ([Zhang et al., 2012](#)) to 50 dB ([Kinos et al., 2016](#)). [Walther et al. \(2017\)](#) draws a theoretical compar-

ison between acousto-optic imaging performances and photoacoustic tomography. This study concluded that acousto-optic imaging is more suited for in-depth imaging as it provides a better contrast to noise ratio. Stefan Kröll and Stefan Andersson-Engels' teams collaborate to simulate an acousto-optic imaging system based on back-scattered signal combined with slow-light detection. This study (Gunther et al., 2018) indicates the promising plausibility of imaging 5 cm of breast tissue in reflection and 12 cm in transmission. The experimental implementation has then been presented by Bengtsson et al. (2019). The authors prove detection up to a 6.8 cm-thick scattering sample.

1.6 Acousto-optic imaging with holographic camera-based detection

In this section we develop the selection and the detection principle of the tagged photons.

1.6.1 Principle of holography

A first common idea to capture light from an object is to use photography. Photography is sensitive to the intensity of light projected on a camera sensor. The conjugation on the sensor with the imaged plane through a lens produces a clear image of the scene. The process is based on the ballistic propagation of light. Then, imaging through a complex scattering medium, where the ballistic regime no longer dominates, requires another approach. Indeed, as the wavefront of the light impinging the camera sensor is largely distorted through the medium, no direct conjugation is possible. In this context, the phase of light must be taken in account to reconstruct the relief of the object, as the phase is linked to the time of flight of light. The discovery of the holography technique in 1949 by Gabor (1949) allowed measuring both the amplitude and the phase of light, hence allowing a reconstruction of the optical field. This discovery was awarded by a Nobel prize in 1971. It is based on the interference between the probe wave and a reference plane wave. Let us denote E_{obj} the field of the probe wave and E_{ref} the field of the reference wave. The light sensor, historically a photographic plate, records the intensity I of the pattern produced by the interference between E_{ref} and E_{obj} . We denote (η, ξ) the set of coordinates in the plane of the camera and (x, y) the set of coordinates in the object plane, or the spatial filter at the output of a scattering sample. Respectively, we denote $\mathcal{E}_{\text{obj}}$ the field in the object plane ($z = 0$) and E_{obj} the field in the camera plane ($z = L$). Between these two planes, the field undergoes a free-space propagation. With the knowledge of the field in one of this two planes we can propagate or back-propagate it numerically through a Fresnel transform — Fourier transform if we neglect the spherical term — to retrieve the field in the other plane. In that case, we have $\mathcal{E}_{\text{obj}} = \tilde{E}_{\text{obj}}$ ($\tilde{E}_{\text{obj}}$ denotes the two-dimensional Fourier transform of E_{obj}). On Figure 1.8, the field in the object plane is reconstructed from the measured complex field in the camera plane¹.

1. In all the thesis, the complex components are represented on figures using an Hue-Saturation-Value (HSV) scale. The hue of each element is given by its phase value and the saturation follows the modulus

The intensity impinging to the camera sensor results from both the contribution of the

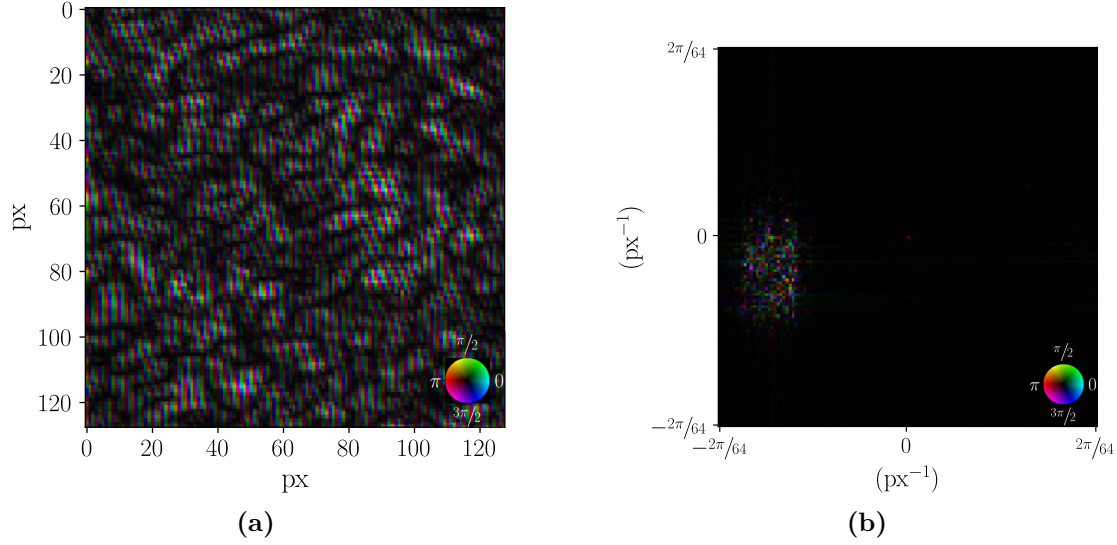


Figure 1.8 – (a) Off-axis heterodyne holographic image of the optical field in the camera plane. The magnitude and the phase of the field are inferred from 4 successive images as we will see below. By performing an inverse two-dimensional Fourier transform on this holographic image, we reconstruct the field in the object plane (b). The edges of the spatial filter placed at the output of the scattering sample (see off-axis scheme on [Figure 1.16](#)) are clearly visible. The image of the filter is located on the left of the Fourier domain due to the off-axis angle between the object and the reference beam.

reference and the object fields. It writes as:

$$I(\eta, \xi) = |E_{\text{ref}} + E_{\text{obj}}(\eta, \xi)|^2, \quad (1.9)$$

$$= |E_{\text{ref}}|^2 + |E_{\text{obj}}(\eta, \xi)|^2 + E_{\text{ref}}^* E_{\text{obj}}(\eta, \xi) + E_{\text{ref}} E_{\text{obj}}^*(\eta, \xi). \quad (1.10)$$

After exposure, the photographic plate must undergo chemical development. For this, one removes it from the setup. The development reveals an optical transmission pattern, denoted $\mathcal{T}$, proportional to the interference intensity received during the exposure. This transmission pattern is then read by the same reference wave and creates three diffraction orders:

$$U(\eta, \xi) = \mathcal{T}(\eta, \xi) E_{\text{ref}}, \quad (1.11)$$

$$\propto I(\eta, \xi) E_{\text{ref}}, \quad (1.12)$$

$$\propto \left(|E_{\text{ref}}|^2 + |E_{\text{obj}}(\eta, \xi)|^2 \right) E_{\text{ref}} + |E_{\text{ref}}|^2 E_{\text{obj}}(\eta, \xi) + E_{\text{ref}} E_{\text{ref}}^* E_{\text{obj}}^*(\eta, \xi). \quad (1.13)$$

of the optical field.

As highlighted by Equation 1.13, the first term, called zero-order, corresponds to the transmitted mean intensities projected in the reference propagation direction. The second term corresponds to the virtual image, order 1. This virtual image is superimposed to the location of the original object. The third one is the real image, projected in the observer side, order -1. In the parallel configuration, the propagation direction of the object and the reference fields are colinear and the three orders are superimposed. Therefore, Leith and Upatnieks (1962) purposed to tilt the reference axis in order to spatially unmix the three images. The so-called off-axis holography thus allows directly reading the virtual and the real images (see Figure 1.9) as they are now spatially separated. In the following decades, development silicon-based electronic devices made the digitization of holography possible. Holograms could be reconstructed from numerically recorded pattern on CCD (Schnars and Jüptner, 1994).

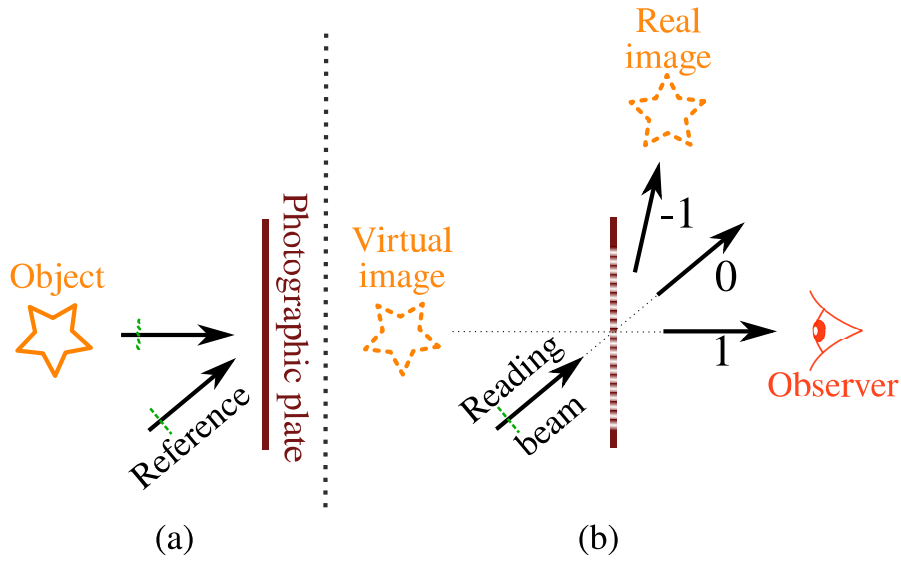


Figure 1.9 – Historical analogic principle of holography in two steps. The first step (a) etches the interference pattern onto the photographic plate. The second so-called reading step (b), the same reference wave is sent through the plate and produce orders of diffraction.

In acousto-optics where the object field mixes optical contribution at different frequencies, off-axis holography can isolate a spectral contribution in order 1. Indeed, the reference field can be shifted in frequency by an acousto-optic modulator to match the frequency of tagged photons. Nevertheless in case of camera detection, as it relies on interferometry, off-axis holography requires to correctly sample the interference fringes. This sampling criterion demands at least to double the number of pixels with respect to Shannon-Nyquist criterion. Then, the correct separation of the diffraction orders limits the angular aperture of the object. With this in mind, Gross et al. (2005a) developed a temporal filtering technique called heterodyne holography.

1.6.2 Temporal filtering

In a parallel detection configuration, the 1-order can however be discriminated from other holographic order by adding a slow temporal modulation to the reading beam. Originally, this slow modulation was produced by a mirror mounted on a piezo-actuator that modifies the optical path. This system has been replaced by two acousto-optic modulators (AOM) that shift the reference beam in frequency (Gross et al., 2003; Atlan et al., 2007). This process allows reducing the noise measurement and avoids the shine of parasitic orders. In the context of acousto-optic imaging, as we have seen in the section 1.4, the beam coming from the object has mainly three spectral components, a central one for the optical carrier and a two side-lobes for positively and negatively-tagged photons, at $\omega_L \pm \omega_{US}$. If we keep the formalism of Equation 1.9, E_{obj} now writes as the sum of three terms corresponding to the untagged photons, at the carrier frequency ω_L , and the two tagged-photons sidebands at $\omega_L \pm \omega_{US}$.

$$E_{obj}(\eta, \xi, t) = E_C(\eta, \xi) e^{i\omega_L t} + E_{M+}(\eta, \xi) e^{i(\omega_L + \omega_{US})t} + E_{M-}(\eta, \xi) e^{i(\omega_L - \omega_{US})t}. \quad (1.14)$$

The limited bandwidth of the camera prevents from detecting the interference fringes of interference between both of the two sidebands. As a consequence, the frequency of the reference beam is shifted at $\omega_L + \omega_{US}$ to match the radiant-frequency of positively-tagged photons². Then to proceed heterodyne holography, the reference is additionally shifted at ω_{mod} . Knowing the frame rate of the camera ω_{cam} , this frequency shift is chosen to be a divisor:

$$\omega_{mod} = \frac{\omega_{cam}}{n}. \quad (1.15)$$

In practice, n is chosen to be equal to 2 or 4, thus we deal with a 2 or 4-phase detection. In total, the reference beam is shifted in frequency at $\omega_{US} + \omega_{cam}/n$.

$$E_{ref}(t) = E_0 e^{i(\omega_L + \omega_{US} + \frac{\omega_{cam}}{n})t}. \quad (1.16)$$

The sensor of the camera, at position $z = L$ (see Figure 1.10), is sensitive to an intensity integrated during a period τ_c :

$$I(\eta, \xi, t) = \int_t^{t+\tau_c} dt' |E_{obj}(\eta, \xi, t') + E_{ref}(\eta, \xi, t')|^2, \quad (1.17)$$

$$\begin{aligned} &= \int_t^{t+\tau_c} dt' \left[|E_0|^2 + |E_C(\eta, \xi, t')|^2 + |E_{M+}(\eta, \xi, t')|^2 + |E_{M-}(\eta, \xi, t')|^2 \right. \\ &\quad + E_0^* E_C e^{-i(\omega_{US} + \frac{\omega_{cam}}{n})t'} + E_C^* E_{M+} e^{i\omega_{US}t'} + \text{c.c.} \\ &\quad + E_C^* E_{M-} e^{-i\omega_{US}t'} + E_0^* E_{M-} e^{-i(2\omega_{US} + \frac{\omega_{cam}}{n})t'} + \text{c.c.} \\ &\quad \left. + E_{M-}^* E_{M+} e^{2i\omega_{US}t'} + E_0^* E_{M+} e^{-i\frac{\omega_{cam}}{n}t'} + \text{c.c.} \right], \end{aligned} \quad (1.18)$$

‘c.c.’ abbreviating the complex conjugate of the previous term in the equation. As the camera integration $\tau_c \gg 2\pi/\omega_{US}$, high-frequency contributions vanish, and thus the signal integrated by the camera is made of:

2. In the textual comments accompanying the equations, we consciously mix up the term of ‘frequency’ denoted f and ‘radiant-frequency’ denoted ω ; $\omega = 2\pi f$.

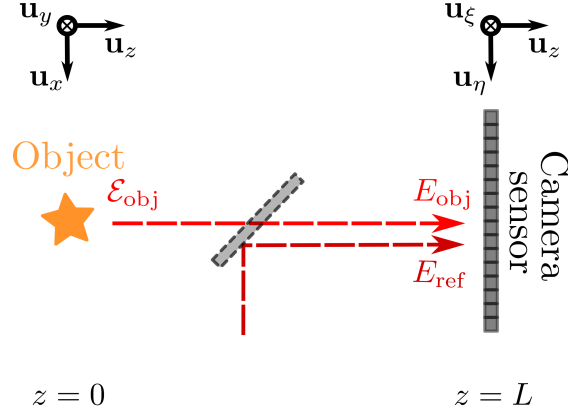


Figure 1.10 – Scheme of the heterodyne holographic detection.

- constant terms corresponding to the sum of the intensities of the two beams, first line in the Equation 1.18, let gather them in a I_0 term,
- a modulation term proportional to the complex amplitude of the positively tagged photons E_{M+} , last line of the Equation 1.18.

We derive this modulation term:

$$\int_t^{t+\tau_c} dt' e^{-i\frac{\omega_{\text{cam}}}{n}t'} = \frac{n}{i\omega_{\text{cam}}} e^{-i\frac{\omega_{\text{cam}}}{n}t} \left(1 - e^{-i\frac{\omega_{\text{cam}}}{n}\tau_c}\right) \quad (1.19)$$

$$= \tau_c e^{-i\frac{\omega_{\text{cam}}}{n}(t+\frac{\tau_c}{2})} \text{sinc}\left(\frac{\omega_{\text{cam}}}{n}\frac{\tau_c}{2}\right). \quad (1.20)$$

with the sinc function $\text{sinc} : t \mapsto \sin(t)/t$. n successive frames I_k are acquired at times $t_k = t_0 + 2\pi k/\omega_{\text{cam}}$:

$$I_k = I_0 + \tau_c \text{sinc}\left(\frac{\omega_{\text{cam}}}{n}\frac{\tau_c}{2}\right) \left(E_0^* E_{M+} e^{-i\frac{\omega_{\text{cam}}}{n}(t_0+\frac{\tau_c}{2})} e^{-2i\pi\frac{k}{n}} + \text{c.c.}\right). \quad (1.21)$$

The n images are numerically combined to reconstruct a complex hologram:

$$H = \sum_{k=0}^{n-1} I_k e^{2i\pi\frac{k}{n}}. \quad (1.22)$$

In 2-phase detection³:

$$H = 2\tau_c \text{sinc}\left(\frac{\omega_{\text{cam}}}{4}\frac{\tau_c}{2}\right) \left(E_0^* E_{M+} e^{-i\frac{\omega_{\text{cam}}}{2}(t_0+\frac{\tau_c}{2})} + \text{c.c.}\right). \quad (1.23)$$

In 4-phase detection:

$$H = 4\tau_c \text{sinc}\left(\frac{\omega_{\text{cam}}}{8}\frac{\tau_c}{2}\right) E_0^* E_{M+} e^{-i\frac{\omega_{\text{cam}}}{4}(t_0+\frac{\tau_c}{2})}. \quad (1.24)$$

3. In this expression we supposed an arbitrary value for the integration time $\tau_c \leq T_{\text{cam}}$ with $T_{\text{cam}} = 2\pi\omega_{\text{cam}}^{-1}$ (successive frame recordings do not overlap). The equation is simplified in the equality case.

In terms of detection quality, the 4-phase detection provides a better signal to noise ratio as it combines two times more measurements. Nevertheless 2-phase detection is more accurate when imaging in an environment where the correlation is quickly lost. Indeed, as it only needs two measurements it is more robust to decorrelation. As we have seen, temporal filtering allows reconstructing the complex contribution of the acousto-optic signals. The field in the object plane can then be retrieved by back-propagating the obtained field through Fresnel or Fourier transform. Because of the colinearity of the two beams in heterodyne holography, the temporal filtering is sensitive to the global intensity variation that affects the numerical reconstruction. As we will see in the next section, spatial filtering allows separating the contribution of fluxes unmatched in frequency.

1.6.3 Spatial filtering

The off-axis holography developed by [Leith and Upatnieks \(1962\)](#) offers a new possibility of filtering. By putting an angle α between the reference and the object beam, this method allows spatially setting apart the real image, the virtual image and the reading beam. A scheme presents the angles between the fields, shown on [Figure 1.11](#). When

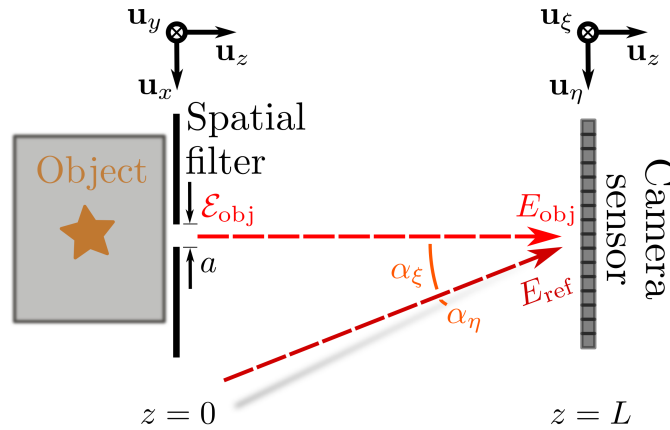


Figure 1.11 – Off-axis schematic of principle with the reference E_{ref} and the object field E_{obj} and the off-axis angles α_η and α_ξ . The spatial filter, a slit of width a , constraints the angular contents of the scattered output field.

we image in scattering media, the scattered field out-coming from the sample contains a large angular contents. In order to limit this content, a diaphragm is placed at the output of the sample, denoted spatial filter on [Figure 1.11](#). The width of this filter will play a role later on. Let us introduce the wave vector of the reference, $\mathbf{k}_{\text{ref}}$:

$$\mathbf{k}_{\text{ref}} = \frac{\omega_L + \omega_{\text{US}}}{c} (\sin(\alpha_\eta) \mathbf{u}_\eta + \cos(\alpha_\eta) \sin(\alpha_\xi) \mathbf{u}_\xi + \cos(\alpha_\eta) \cos(\alpha_\xi) \mathbf{u}_z). \quad (1.25)$$

Supposing the reference beam has a flat wavefront close to the camera plane $z = L$, the reference field writes as:

$$E_{\text{ref}}(\eta, \xi, z, t) = E_0 e^{-i \frac{\omega_L + \omega_{\text{US}}}{c} (\sin(\alpha_\eta) \eta + \cos(\alpha_\eta) \sin(\alpha_\xi) \xi + \cos(\alpha_\eta) \cos(\alpha_\xi) z)} e^{i(\omega_L + \omega_{\text{US}} + \frac{\omega_{\text{cam}}}{n})t}, \quad (1.26)$$

with E_0 a constant. For the simplicity, we consider $\alpha_\eta \ll 1$ and $\alpha_\xi \ll 1$, this approximation will be justified in section describing the separation criterion in the Fourier domain.

$$E_{\text{ref}}(\eta, \xi, z, t) = E_0 e^{-i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + z)} e^{i(\omega_L + \omega_{\text{US}} + \frac{\omega_{\text{cam}}}{n})t}. \quad (1.27)$$

As we wrote in the previous section, the sensor of the camera, located at $z = L$ integrates the following intensity:

$$\begin{aligned} I(\eta, \xi, L, t) = \int_t^{t+\tau_c} dt' & \left[|E_0|^2 + |E_C|^2 + |E_{M+}|^2 + |E_{M-}|^2 \right. \\ & + E_0^* e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} E_C e^{-i(\omega_{\text{US}} + \frac{\omega_{\text{cam}}}{n})t'} + \text{c.c.} \\ & + E_0^* e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} E_{M+} e^{-i \frac{\omega_{\text{cam}}}{n} t'} + \text{c.c.} \\ & + E_0^* e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} E_{M-} e^{-i(2\omega_{\text{US}} + \frac{\omega_{\text{cam}}}{n})t'} + \text{c.c.} \\ & + E_C^* E_{M+} e^{i \omega_{\text{US}} t'} + E_C^* E_{M-} e^{-i \omega_{\text{US}} t'} + \text{c.c.} \\ & \left. + E_{M-}^* E_{M+} e^{2i \omega_{\text{US}} t'} + \text{c.c.} \right]. \end{aligned} \quad (1.28)$$

As $\tau_c \gg 2\pi/\omega_{\text{US}}$, high-frequency terms do not contribute to the recorded intensity. The contribution of positively tagged photons appears as intensity fringes superimposed on a background formed by the intensity of untagged or opposite frequency-shifted photons and reference. In standard holography, the separation between order 0 and ± 1 occurs when the etched hologram is read by the reference beam. The hologram inscription in the photographic plate is mathematically described by the Fresnel-Kirchhoff integral (Schnars and Jüptner, 2002). Under the Fresnel approximation and except a spherical term, the object field reconstruction is similar to an inverse two-dimensional Fourier transform of the hologram. With the development of CCD, the recording and reading of the interference pattern have been digitized. The two-dimensional Fourier transform (FT2D) links the object to the hologram. We define the FT2D of the field E in the plane of coordinate (η, ξ) as:

$$\tilde{E}(\nu_\eta, \nu_\xi) = \int_{\mathbb{R}^2} d\eta d\xi E(\eta, \xi) e^{-2i\pi(\nu_\eta \eta + \nu_\xi \xi)}. \quad (1.29)$$

We denote $\nu_\eta^0 = \frac{f_L + f_{\text{US}}}{c} \alpha_\eta$ and $\nu_\xi^0 = \frac{f_L + f_{\text{US}}}{c} \alpha_\xi$.

The FT2D of the intensity pattern of Equation 1.28 also writes as:

$$\begin{aligned} \tilde{I}(\nu_\eta, \nu_\xi, L, t) = \tau_c \Big\{ & I_0 \delta(\nu_\eta, \nu_\xi, L) + \left(\tilde{I}_C + \tilde{I}_{M+} + \tilde{I}_{M-} \right) (\nu_\eta, \nu_\xi, L) + \text{sinc} \left(\frac{\omega_{\text{cam}} \tau_c}{n} \frac{\tau_c}{2} \right) \times \\ & \left[E_0^* e^{-i \frac{\omega_{\text{cam}}}{n} (t_0 + \frac{\tau_c}{2})} \tilde{E}_{M+}(\nu_\eta, \nu_\xi, L) * \delta(\nu_\eta - \nu_\eta^0, \nu_\xi - \nu_\xi^0) \right. \\ & \left. + E_0 e^{i \frac{\omega_{\text{cam}}}{n} (t_0 + \frac{\tau_c}{2})} \tilde{E}_{M+}^*(\nu_\eta, \nu_\xi, L) * \delta(\nu_\eta + \nu_\eta^0, \nu_\xi + \nu_\xi^0) \right] \Big\}, \end{aligned} \quad (1.30)$$

with δ the Dirac function. The distribution of the Fourier components in the two-dimensional Fourier domain (Figure 1.12) shows a central spike corresponding to the Dirac contribution $\delta(\nu_\eta, \nu_\xi, L)$ in the Equation 1.30. This central spike is surrounded by a large spot identified to the sum of the FT2D $\tilde{I}_C + \tilde{I}_{M+} + \tilde{I}_{M-}$. Writing this contribution in the form:

$$\tilde{I}_C(\nu_\eta, \nu_\xi, L) = \widetilde{E_C^* E_C}(\nu_\eta, \nu_\xi, L), \quad (1.31)$$

$$= \left(\tilde{E}_C^* * \tilde{E}_C \right) (\nu_\eta, \nu_\xi, L), \quad (1.32)$$

highlights the triangular shape of the spot created by the convolutions $\tilde{E}_C^* * \tilde{E}_C$, $\tilde{E}_{M+}^* * \tilde{E}_{M+}$ and $\tilde{E}_{M-}^* * \tilde{E}_{M-}$. Finally two symmetrical lobes proportional to the FT2D of the optical tagged field in the camera plane, second and third lines of Equation 1.30, are separated from the center (Gross et al., 2008). As we see on Figure 1.12 the off-axis holography spatially separates the contribution of tagged photons from the main untagged light in the Fourier domain. With the 2-phase detection we get rid of the central spot and keep the symmetrical and complex conjugate lobes, following Equation 1.22:

$$\begin{aligned} \tilde{I}(\nu_\eta, \nu_\xi, L, t) = 2\tau_c \text{sinc} \left(\frac{\omega_{\text{cam}} \tau_c}{4} \right) \times \\ \left[E_0^* e^{-i \frac{\omega_{\text{cam}}}{2} (t_0 + \frac{\tau_c}{2})} \tilde{E}_{M+}(\nu_\eta, \nu_\xi, L) * \delta(\nu_\eta - \nu_\eta^0, \nu_\xi - \nu_\xi^0) \right. \\ \left. + E_0 e^{i \frac{\omega_{\text{cam}}}{2} (t_0 + \frac{\tau_c}{2})} \tilde{E}_{M+}^*(\nu_\eta, \nu_\xi, L) * \delta(\nu_\eta + \nu_\eta^0, \nu_\xi + \nu_\xi^0) \right]. \end{aligned} \quad (1.33)$$

The four-phase detection then extracts only one of the two lobes and refines the SNR (Gross et al., 2008):

$$\tilde{I} = 4\tau_c \text{sinc} \left(\frac{\omega_{\text{cam}} \tau_c}{8} \right) E_0^* e^{-i \frac{\omega_{\text{cam}}}{4} (t_0 + \frac{\tau_c}{2})} \tilde{E}_{M+}(\nu_\eta, \nu_\xi, L) * \delta(\nu_\eta - \nu_\eta^0, \nu_\xi - \nu_\xi^0). \quad (1.34)$$

Figure 1.13 shows the corresponding 2 and 4-phase Fourier domain. As seen in the Equation 1.33, the 2-phase detection removes the central component, leaving the two symmetrical lobes corresponding the two complex conjugates, proportional to the acousto-optically tagged field. The Equation 1.34 only keeps one of these two lobes. Once we have separated the tagged light contributions in the Fourier domain, we can select the Fourier components of the tagged light to retrieve the complex tagged field as it impinges the camera sensor surface. For this we crop the Fourier domain around the lobe and

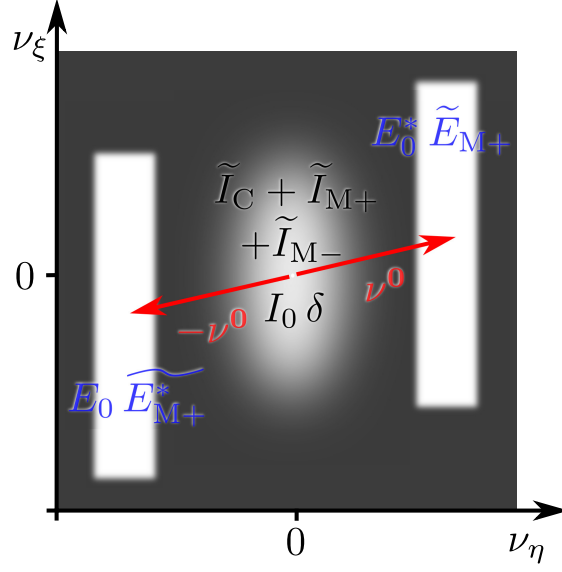


Figure 1.12 – Scheme of the order separation in off-axis holography. As written in the Equation 1.30 after the FT2D, the $\tilde{E}_{M+}$ and $\tilde{E}_{M+}^*$ contributions are symmetrically shifted outside of the central spot. $\nu^0 = (\nu_\eta^0, \nu_\xi^0)$ is directly linked to the off-axis tilt angles α_η and α_ξ depicted on Figure 1.11.

center it on a empty Fourier domain (see Figure 1.14). Re-centering the Fourier components removes the phase slope remaining from the off-axis angle. Once re-centered an inverse FT2D builds us the complex acousto-optic field in the camera plane. This knowledge of the complex field gives us access to the phase of the optical wave. As we see in forthcoming sections, measuring the phase of the acousto-optically tagged field will allow us to perform phase manipulation and wavefront shaping.

In practice, intensity fluctuations along the measurement diminish the efficiency of extraction. In particular, some components remain in the center of the spectral domain at low frequency. For instance the fast camera that we use for measuring the acousto-optic transmission matrix (see chapter 3), has a window in front of its sensor. This thin glass layer creates a low spatial modulation on all the recorded images. In the spatial Fourier domain, it manifests through two symmetrical spots at the center of the domain. Also, on the Fourier-transform acousto-optic imaging setup (see chapter 2), some reflections on a mirror mount creates some parasitic artifacts at the center of the Fourier domain. We got rid of it by isolating the mount with black mat metallic sheets. In order to collect only the contribution of the tagged photons, spectral orders need to be clearly distinct with no overlap. To that extent a spatial filter, represented on Figure 1.11, is placed at the output of the scattering sample in the (x, y) plane. The shape and the aperture of this diaphragm have to be chosen with respect to the following criteria implying the distance between the diaphragm and the camera sensor L , and the pixel pitch Δ_{pix} . The diaphragm consists in a slit of width a and supposed to be infinite

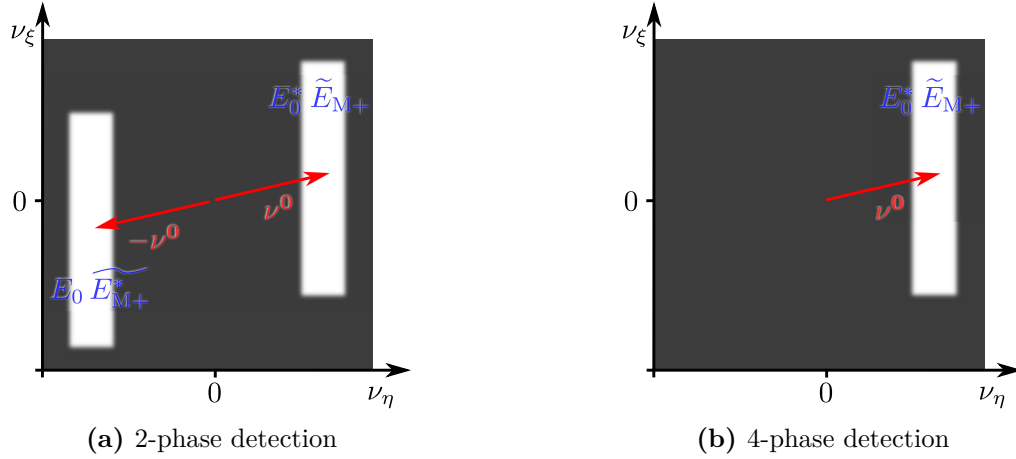


Figure 1.13 – Illustration of the result of 2-phase (a) and 4-phase (b) combination in the spatial Fourier domain. On both of these figures, the linear combination of the images suppress the central component. 2-phase keeps the two symmetric lobes proportional to the acousto-optically tagged field and its complex conjugate. As 4-phase detection adds a supplemental combination along the imaginary part, it only keeps one of these two lobes.

in the second direction. The reference beam is focused through a lens in the plane of the spatial filter, see [Figure 1.16](#). The focal distance of the lens is chosen to focus in the plane of the diaphragm and uniformly covers the surface of the camera sensor.

Sampling the speckle For this, speckle grains have to be spread enough to overlap at least 2 pixels of the camera sensor. As seen with [Equation 1.4](#), the speckle size δ_η is given, under the Fraunhofer far-field approximation $\frac{a^2}{\lambda L} \ll 1$, by:

$$\delta_\eta = \frac{\lambda}{\text{NA}} = \frac{\lambda L}{a}. \quad (1.35)$$

This gives the constraint:

$$\delta_\eta = \frac{\lambda}{\text{NA}} = \frac{\lambda L}{a}. \quad (1.36)$$

Sampling the fringes In order to be correctly sampled by the camera sensor, maximum and minimum intensity on the interference fringes need to spread at least on two different pixels. This imposes:

$$\alpha_\xi \leq \frac{\lambda}{2\Delta_{\text{pix}}}. \quad (1.37)$$

Separate off-axis orders To be distinguishable in the Fourier domain from the central spot of width $2a$, the lobe of width a their centers have to been separated at least

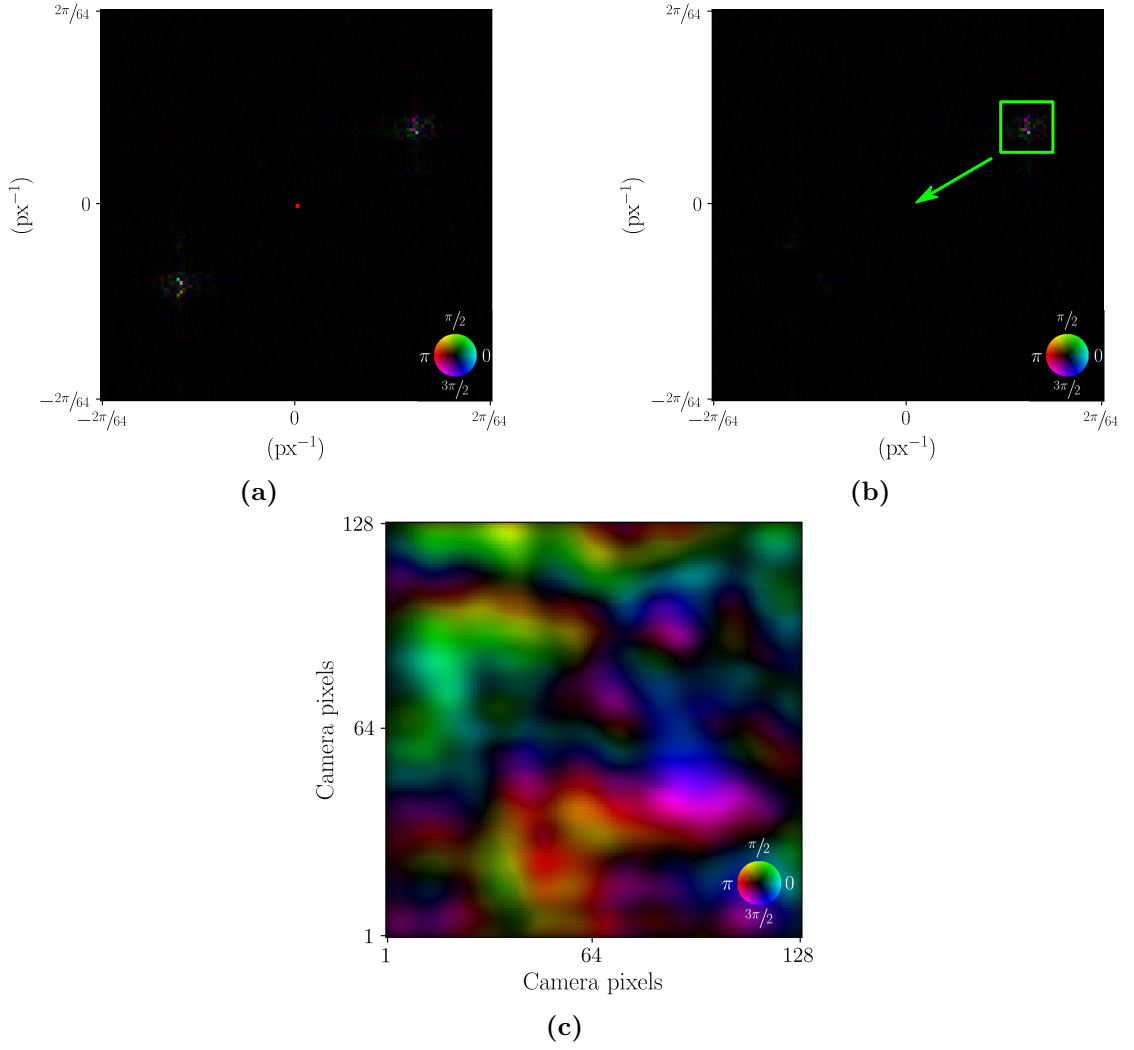


Figure 1.14 – (a) Spatial Fourier domain for off-axis 4-phase heterodyne detection. We see the acousto-optic contributions and remaining contributions of the central order. Isolating the lobe, re-center it in the Fourier domain (b) and processing an inverse two-dimensional Fourier transform, we calculate the acousto-optic speckle in the sensor plane (c).

with $3a/2$ (see Figure 1.15) so:

$$\alpha_\xi L \geq \frac{3a}{2}. \quad (1.38)$$

The wavelength of our laser source and the pitch of the pixels of our camera are:

$$\lambda = 780 \text{ nm} \quad \Delta_{\text{pix}} = 13.6 \mu\text{m}. \quad (1.39)$$

Experimentally, the width of the slot and the distance to the camera are chosen first to ensure the sampling of speckle grains with $L = 60 \text{ cm}$ and $a = 8 \text{ mm}$. The α angle is

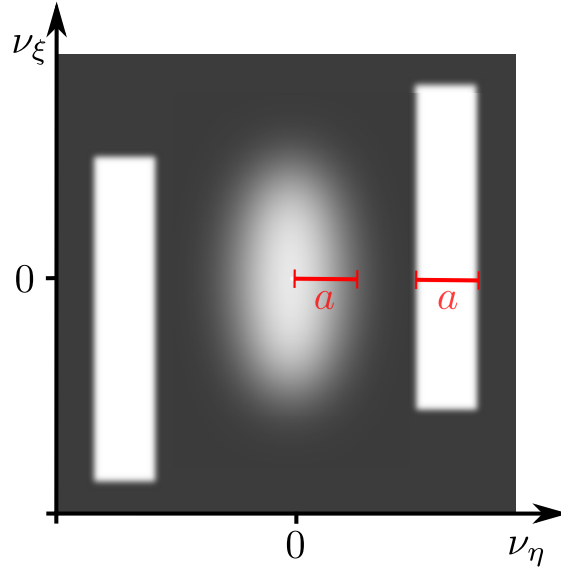


Figure 1.15 – Slot width correspondence in the spatial Fourier domain.

tuned using to mirrors (far-field, near-field) to adjust the off-axis lobes position in the Fourier domain, ideally halfway between the origin and the edge. For this two programs have been developed for each of the two setups presented in the following chapters respectively in Labview and Python respectively. These programs trigger and process the multiple-phase detection in real time. This development brought ease for fine tuning the off-axis detection on the optical setup while keeping an eye on the reconstruction in the spatial Fourier domain. Once the optimal angle is found, the width of the slot is adjusted to benefit from an optimal area in the Fourier domain.

The [Figure 1.16](#) presents the off-axis detection optical setup. The slot is made from two copper plates mounted on a 3D printed polylactic acid (PLA) frame. This configuration ensures the stability of the spatial filter. Indeed, as we will see in the [chapter 3](#) the stability of the spatial correlation of the reconstructed AO field which is a necessary condition for measuring the acousto-optic transmission matrix of a scattering sample. As the reference wave is focused in the slot plane, we consider them as two interfering source points coming from the same plane.

1.6.4 Extracting the signal of tagged photons

As we have seen, the signal intensity of the lobe in the Fourier domain is proportional to the 2D Fourier transform of the tagged light. Experimentally we use a fast Fourier transform algorithm to compute the FT2D. According to the Parseval's equality, the integrated intensity of the complex hologram H defined in [Equation 1.22](#) on the off-axis lobes is:

$$s^\phi = \langle |H|^2 \rangle \propto |\tilde{E}_{\text{obj}}|^2. \quad (1.40)$$

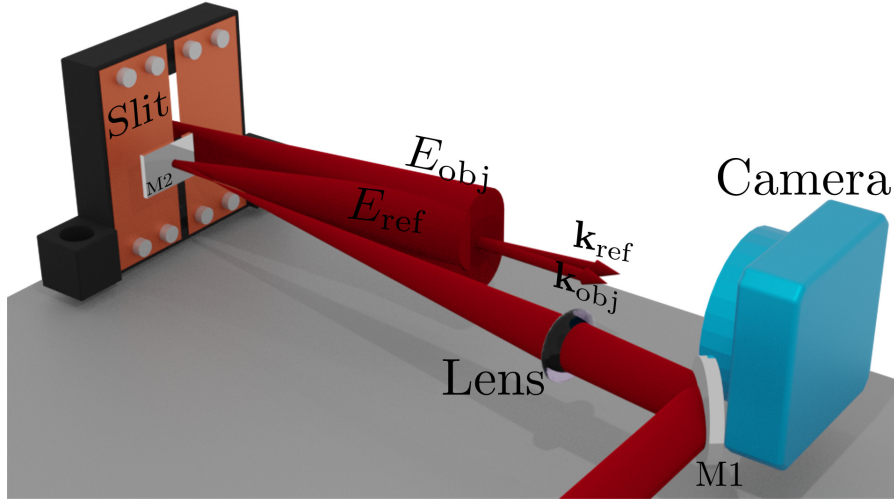


Figure 1.16 – Scheme of the off-axis detection setup. The slot is made from two copper plates mounted on a 3D printed frame. This configuration ensures the stability of the spatial filter. Mirrors M1 and M2 allow finely tuning the angle of the reference wave and as a consequence the position of the off-axis symmetrical lobes in the Fourier domain. The lens focuses the reference wave in the slit plane (conjugated by the mirror M1).

This quantity is proportional to the acousto-optically tagged intensity recorded on the integration time of the camera, which typically is of the order of 10 to 100 μs depending on the temporal modulation applied on the ultrasound. This signal s can be normalized by the noise in the remaining area of the Fourier domain by:

$$s_N = \frac{s - s_0}{s_0}. \quad (1.41)$$

where s_0 is the integrated intensity on a free area of equal surface in the Fourier domain. Indeed the fluctuations in this area correspond to the TF2D of the noise measurement of the camera. The camera records the intensity signal on gray levels encoded on 10 bits, i.e. 1024 levels.

1.7 Conclusion

In this chapter, we have seen why optical imaging techniques are crucial for the diagnosis and early detection of some diseases. After developing the reason why multiple scattering imposes a limit to standard optical imaging techniques, we have shown that the complex scattered pattern emerging by shining such a medium still contains information on light source and its history. The acousto-optic effect allows tagging light passing at a specified region. The amplitude of the tagged light is directly linked to the local contrast due to absorption and scattering. Finally, we have presented digital holography which

allows specifically the detection of the acousto-optic signal. All these elements gathered constitute acousto-optic imaging in a camera-based detection.

CHAPTER 2

Shaping the ultrasonic excitation

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IN this chapter, we develop the acousto-optic tagging modalities used in combination with off-axis holographic detection. As the core of acousto-optic imaging relies on the propagation of the ultrasounds in the imaged zone, we first focus on the temporal modulation. In this first step we will describe techniques using a simple acoustic probe providing a spatially focused wave. Secondly we will present designed spatial modulations used to image with a wider field of tagging. This wave shaping technique requires the use of a multi-channel probe controlled by an arbitrary waveform generator.

2.1 Temporally modulated ultrasounds

The most common and basic scheme to perform acousto-optic imaging is to use spatially-focused ultrasounds for insonification. In this configuration, the path of acoustic wave is guided by the geometry of the emitters through a column-shape volume across the medium. The containment of the acoustic wave thus offers a primary localisation in the volume to be imaged in the plane transverse to the propagating direction. Once placed on a motorized stage, we scan the excitation across the volume of interest point by point.

A simple way to produce these focused waves is to use spherical ultrasonic transducer. The geometry of the ultrasonic probe guaranties the focusing of the ultrasonic wave at a focal point. It consists in a spherical membrane attached on a piezoelectric actuator. The simplicity of these devices makes them mechanically robust enough to be installed on a motorization stage while reaching the megapascal order of magnitude for pressure. The signal they emit are controlled by a function generator, or an arbitrary waveform generator for complex modulated signals, and magnified by a power amplifier. Furthermore, a range of spherical single-element transducers are commercialized for multiple spectral bandwidths and multiple central-emission frequencies. For example we own few of these transducers at 2 MHz, 5 MHz, 15 MHz and 25 MHz. The design of these probes imposes nevertheless a determined focusing distance. In this work we commonly used a 5 MHz probe whose focusing distance is 50 mm for an element-width of 25 mm, which gives us a 0.5 numerical aperture.

We measured the profile of pressure emitted by the probe quantitatively on a calibrated measuring bench at the BioMaps¹ laboratory with Jean-Luc Gennisson and Anthony Novell. These pressure maps presented on [Figure 2.1](#) were acquired by scanning a piezo-electric ultrasonic sensor on a three axis translation stage. The emissions were repeated for each positions. A controlled delay for triggering the oscilloscope allows capturing the excitation after the desired time of flight. The excitation consisted in pulses of dozen of acoustic periods. This temporal length allows measuring the peak-to-peak pressures away from any visible edge effect. These peak-to-peak measurements are presented on [Figure 2.2](#). They exhibit a cigar-shaped acoustic spot. Indeed, [Figure 2.2\(a\)](#) reveals an axial extension on 10 mm compared to the transverse profile of 0.75 mm width at half-maximum. The extent of the acoustic map is inherent to the

1. Laboratory of multi-modal biomedical imaging in Paris-Saclay

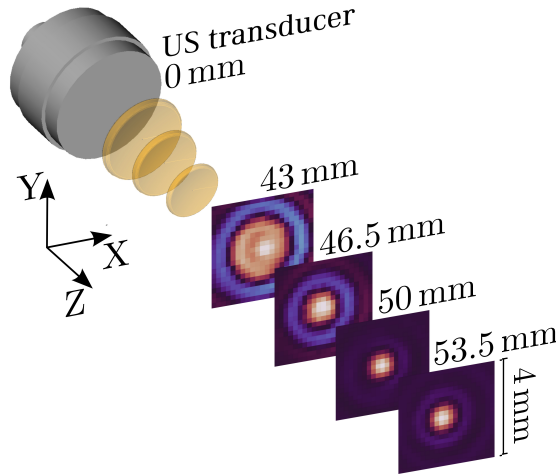


Figure 2.1 – Scheme of the spherical single-element 5 MHz probe whose focal distance is 50 mm. Four experimentally-measured pressure maps are displayed along the acoustic propagation axis.

geometry of the emitting probe. Moreover, the imaging resolution is given by the interaction zone between the optical and the acoustic wave during the integration time of the camera. Although, short pulsed excitations appear as a simple solution.

2.1.1 Brief integration time

As we have seen in the first chapter, the resolution of the optical contrast probed by measuring the signal from the acousto-optic interaction is given by the intersection of the focusing area of the ultrasonic probe and the spread of the scattered optical beam. But the acoustic focusing zone spreads on few millimeters for probes in the megahertz range offering a coarse resolution. Then, the duration of collection by the camera adds a controllable selection of the tagged signal for example during the propagation of the acoustic wave. In order to temporally gate the signal, we use a brief integration time, i.e. few microseconds. We combine this brief detection with temporal modulation of the acoustic wave to provide spatial sectioning along the propagation.

Pulsed ultrasounds

A first idea to section the tagged photons along the acoustic propagation axis is to use pulsed ultrasounds. The fast camera then allows capturing the photons tagged during the propagation of the ultrasounds. As the acoustic propagation is predominantly ballistic in the megahertz range in biological tissue, the insonification of the medium is controllable. Nevertheless, the volume of this area in the medium is proportional to the length of the acoustic pulse, the waist of the acoustic column and the integration time. As a result, improving the resolution by reducing the temporal length of pulses comes

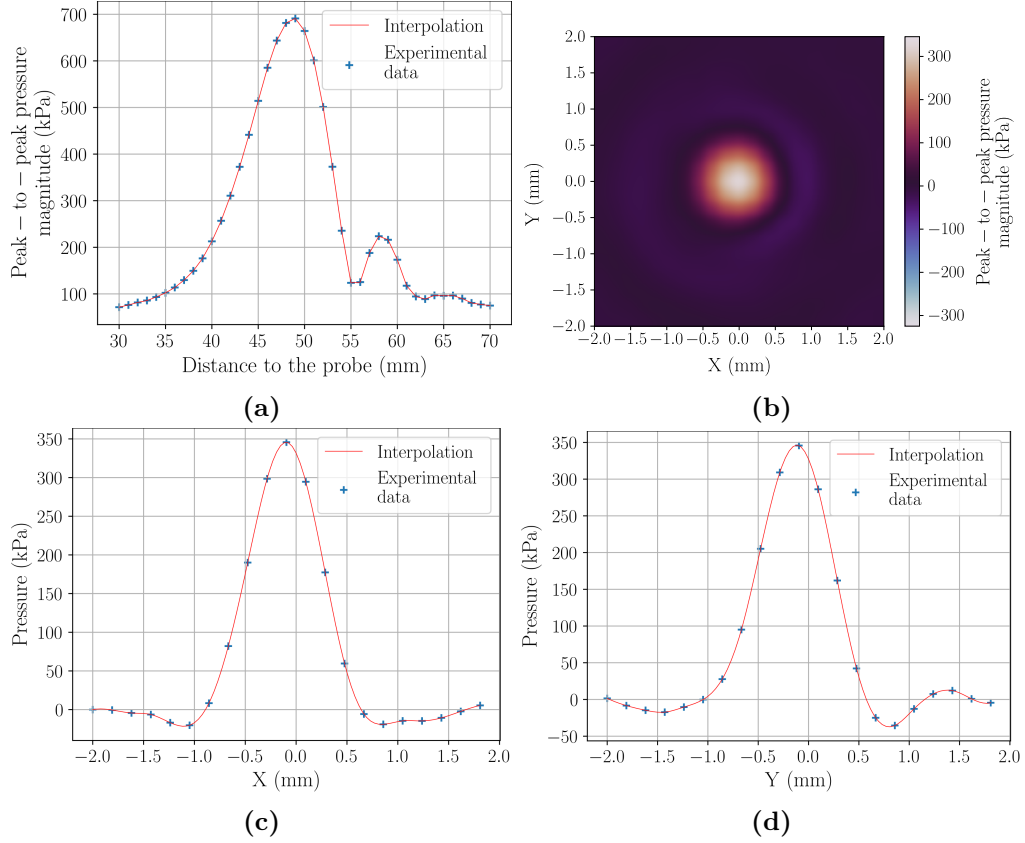


Figure 2.2 – Pressure profile of the 5 MHz ultrasonic spherical transducer used for the AOTM measurement. The axes X , Y and Z correspond to those of Figure 2.1. (a) Axial pressure profile (along the propagation axis). (b) Transverse pressure map at the focus position at 50 mm. (c) and (d) are the transverse pressure profiles along X and Y axes display on (b).

along with reducing the quantity of tagged photons. Then we need to average the measurements hundreds of times to maintain a significant signal-to-noise ratio. Furthermore the briefness of the pulses implies putting more pressure power in the medium. In the context of biomedical imaging this method is constrained by acoustic-exposition norms (Phillips and Harris, 2008).

In order to scan axially along the ultrasound propagation axis, a delay between the pulse generator and the camera integration has to be precisely controlled and repeated to correctly accumulate the measurements for averaging Atlan (2005). To illustrate this principle, we present on Figure 2.3 a sequence of successive images taken across the propagation of a 2 MHz centered ultrasonic pulse. The hydrogel imaged in this experiment is made of agar 4 % and two inclusions of absorbed black ink buried inside. The agar recipes allows producing a hydrogel with a Young’s modulus similar to mechanical properties of soft biological tissues, 50 kPa. Hence, like in biological tissue, we can con-

sider the propagation of acoustic waves as harmonic. Only using agar in the gel recipes makes a limpid sample in which light undergoes ballistic propagation. This is useful for rapid detection diagnosis and to prove the principle of the experiment before moving to highly scattering samples.

2.1.2 Experimental example of sampling the pulse propagation with a camera

As an example on [Figure 2.3](#) we recorded a movie of the propagation of spatially focused 3 MHz ultrasonic pulse across two ink inclusions trapped in a limpid hydrogel. Here, as we collect ballistic illumination, the insonified plane can be optically conjugated

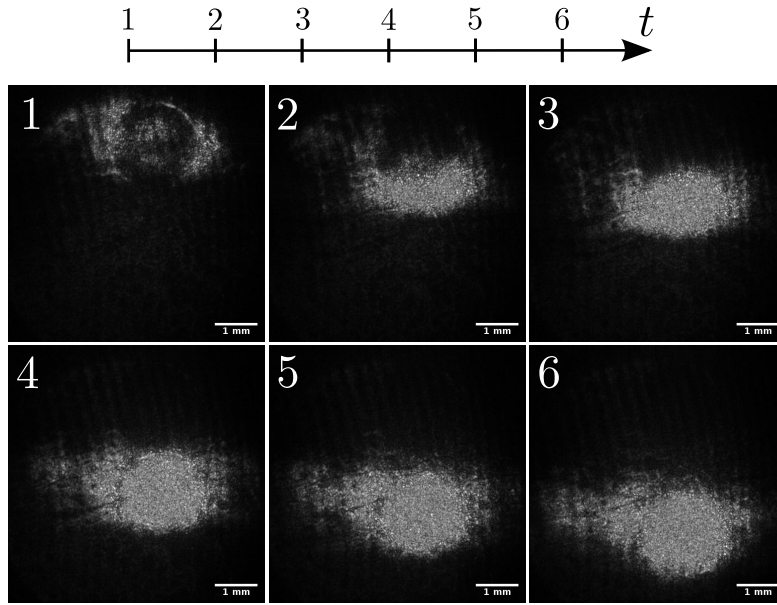


Figure 2.3 – Propagation of an ultrasonic pulse at 3 MHz through a limpid hydrogel. The movement of the pulse is captured using off-axis heterodyne detection. The two first images show the shadow of the black-ink inclusion buried into the sample. The absorption of the inclusion kills the acousto-optic signal.

with the camera's sensor plane (see [Figure 2.4](#)). In this configuration the resolution of the imaging in the propagation direction is ruled by the width of the pulse and the integration time of the camera. Indeed, the light tagged during the temporal length of the acoustic pulse is captured in the integration time of the camera. As a consequence, using this technique of modulation, improving the resolution walks aside with reducing the collected signal. Even if this is possible in ballistic imaging, the weak signal reaching out of high scattering samples puts a limit to this strategy.

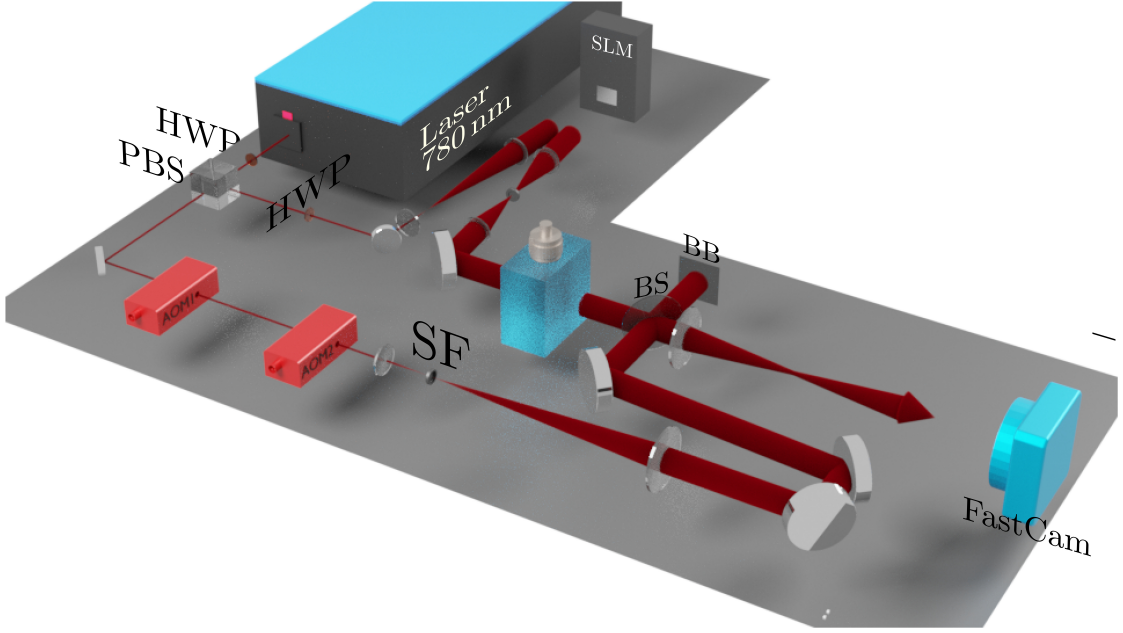


Figure 2.4 – Optical setup used to image the insonified plane collecting the ballistic light. AOM# for acousto-optic modulators, HWP half-wave plates, SF spatial filter (pin-hole) of diameter $200\ \mu\text{m}$, (P)BS (polarized) beam splitter, SLM spatial light modulator, simply used without modulation here, equivalent to a mirror. BB for beam block. A lens conjugates the ultrasound plane to the sensor of the camera plane. As imaging is ballistic, a spatial filter in the output surface of the sample is no longer needed. The reference beam is directed toward the camera (FastCam) by a beam-splitter plate. The off-axis angle is tuned by tilting the beam-splitter and the upstream mirror.

Frequency-swept acoustic pulses

Another method proposed by Yao and Wang (2000) and experimentally set up in the aftermath in Yao et al. (2000) is based on a linear sweep of frequency, namely chirp. This configuration relies on light beam illuminating the scattering sample. The frequency of the ultrasounds f_{obj} is swept following the law:

$$f_{\text{obj}}(t, \tau) = f_{\text{US}} + b(t - \tau), \quad (2.1)$$

with b the slope of the frequency ramp. The temporal delay is denoted τ , f_{US} is the central frequency of the ultrasound. The ultrasonic wave is also modulated by a sinusoidal amplitude. By phase-shifting this modulation, the authors separate the tagged-photon contribution from the untagged light that is not modulated. As a result, for a given delay τ the camera acquires four successive images. The camera bandwidth rejects the tagged photons coming from outside the region corresponding to the time of flight τ of

the ultrasounds in the acoustic propagation column.

This method was put further by [Forget et al. \(2003\)](#). The light source is also modulated in amplitude by a chirp:

$$\cos [2\pi (f_1 + bt)]. \quad (2.2)$$

The slope of the frequency sweep b is the same as the one that modulates the ultrasounds. Along the propagation of the ultrasonics the pressure field writes as:

$$P(x, y, z, t) = P_0 A(x, y, z, t) \cos \left[2\pi (f_2 + bt) t - 2\pi \frac{f_2 + bt}{c_s} z \right], \quad (2.3)$$

with c_s the celerity of sound in the medium, and $A(x, y, t)$ the envelops of the acoustic field depending on the geometry of the probe in ballistic regime. As a result, the tagged light received by the camera undergoes slow fluctuations of amplitude, high-frequencies are averaged by the integration of the camera:

$$\cos \left\{ 2\pi \left[f_1 - f_2 + \frac{bz}{c_s} \right] t + 2\pi \frac{f_2 z}{c_s} \right\}. \quad (2.4)$$

The slight frequency mismatch, $f_1 - f_2$ in the cosine function, plays the role of a delay which can be tuned in order to select the desired depth z . The frequency of this slow modulation follows a frequency sweep linked to the progression of the acoustic wave across the medium. By matching the pixels of a fast camera with speckle grains of the output field, they record a movie of the optical flutter produced by the chirp. Because of the propagation and the knowledge the acoustic wave, they linked the Fourier spectrum of this flutter to the virtual acousto-optic sources locations. Although one movie gives a direct access to the axial acousto-optic profile. [Atlan \(2005\)](#) thus reduced down to 70 ms the time of one line acquisition through a thick sample, whose thickness was around 40 to 50 times transport mean free path. Despite this progress the method remains sensitive to decorrelation in living tissues.

Temporal modulation of the acoustic wave comes as a natural solution in order to improve the resolution along the propagation axis.

2.1.3 Long duration pulsed chirp

Chirp is a modulation used for example by [Yao et al. \(2000\)](#). It consists in a long pulse modulated in phase using a temporal ramp of frequency. At the Institut Langevin, long chirp was used by [Forget et al. \(2003\)](#) to image ink inclusions in 5 mm of diameter whose centers were separated by 8.5 mm. Supposing that the camera continuously acquires N frames at the rate f_{cam} , the spatial resolution is given by:

$$\Delta z = \frac{c_s}{b} \Delta f \quad \text{with} \quad \Delta f = \frac{f_{\text{cam}}}{N}. \quad (2.5)$$

As a result, the resolution relies on the shortness and the frame rate of the high speed camera. [Forget et al. \(2003\)](#)'s work indicates a resolution of 1.2 mm for a camera rate of 200 Hz and 256 images.

2.1.4 Long integration time using autocorrelated signals

In order to maximize the collected signal, [Lesaffre et al. \(2009\)](#) proposed to use long duration complex waveforms. The purpose of this method, namely acousto-optic coherent tomography, consists in controlling the optical coherence between the object and reference beams using a common modulating signal. The integration provided by the camera sensor during the exposure time is used as an analogical autocorrelation operation between the two modulations applied on the two arms. Indeed, as we use a light source of long coherence time, both arms stay correlated along the integration. Wisely choosing the correlation properties by shaping of the modulation signal we can select an identified area along the ultrasonic propagation axis. Moreover the coherence allows to integrate the speckle longer and thus collect more acousto-optic signal. The two modulations has to be synchronized in time to temporally match when they meet the sensor of the camera. In the following section we present two types of modulations. The first one, random phase jumps, used coherence property of random signals. The second is based on chirp signal but used for its autocorrelation property.

Random phase jumps

The principle of modulating the optical beams by a random-phase-jump pattern was first imagined by [Lesaffre et al. \(2009\)](#). The experimental technique exploiting the properties of such random signals was then implemented in the aftermath at the Langevin Institute by [Lesaffre et al. \(2011\)](#) in combination with an holographic setup using a photorefractive crystal. This work was followed by ([Benoit à la Guillaume et al., 2012](#)) with an off-axis holographic detection. It consists in modulating a long carrier pulse by a sequence of random -1 and 1 periodically distributed. The period of each $-1, 1$ modulation is equal to T_0 , about a microsecond, the same order of magnitude that for pulses. As we will see later in this section, the width of the jumps determines the resolution of the method. We call it random phase jumps as multiplying by -1 or 1 is equivalent to π -shifting the phase or not. The total length of the random pattern is given by $N T_0$ the total number of jumps. This modulation conserves the power of the pulse as playing only with the phase of the wave. The randomness of the modulation pattern guaranties its non-periodicity, which would affect the localization of the virtual acousto-optic sources as we will see later. The same generated random pattern is applied on both the ultrasonic wave and the reference beam through the second acousto-optic modulator (see [Figure 2.5](#)). One of the two modulations is applied with a specific delay to select a depth along the ultrasound propagation.

Let us denote $\rho(t)$ the random-phase-jump pattern built with N jumps of duration T_0/N :

$$\rho(t) = \sum_{k=0}^{N-1} b_k \Pi\left(t - \frac{k}{N}T_0\right) \quad \Pi(t) = \begin{cases} 1 & \text{if } -T_0/2N \leq t < T_0/2N \\ 0 & \text{otherwise} \end{cases}, \quad (2.6)$$

with b_k randomly equals to 1 or -1 . [Figure 2.6](#) presents a realization of the modulation pattern ρ .

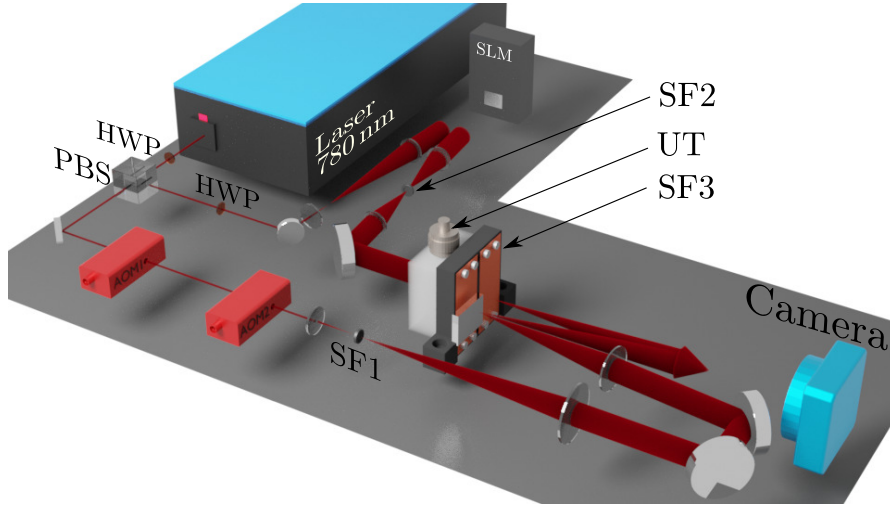


Figure 2.5 – Optical scheme of the setup used for imaging. At this stage, the spatial light modulator is simply used as a mirror and so has no action on the setup. The modulation is played by the second acousto-optic modulator (AOM) and the ultrasonic transducer (UT). We used a multiple scattering hydrogel as sample.

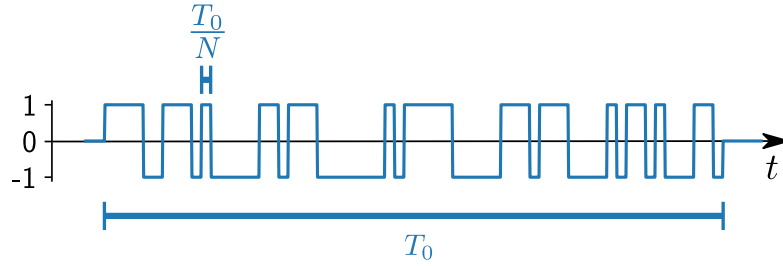


Figure 2.6 – Random modulation pattern $\rho(t)$ modulating the ultrasound signal and the reference beam. The signal jumps T_0 periodically to -1 or 1 (0 or π in term of phase). The order of magnitude of the temporal width T_0 of each phase jump is the microsecond. As the total number of phase jumps is N , the temporal length of the pattern is $N T_0$.

The modulated pressure wave emitted by the transducer is:

$$P(t) = P_0 \rho(t) e^{i\omega_{\text{US}} t}, \quad (2.7)$$

where P_0 denotes the nominal pressure. Along the ultrasound propagation axis y , by neglecting diffraction effects, we write the pressure wave within the medium as:

$$P(y, t) = P_0 \rho(t - c_s^{-1} y) e^{i\omega_{\text{US}} (t - c_s^{-1} y)}. \quad (2.8)$$

The reference beam used for off-axis heterodyne detection is shifted in frequency at $\omega_L + \omega_{\text{US}}$ in order to select one sideband of the tagged photons. It is also modulated

by the random pattern and has the following expression with respect to the notation introduced in the [subsection 1.6.3](#) developing spatial filtering in off-axis holographic detection:

$$E_{\text{ref } \tau}(\eta, \xi, L, t) = E_0 \rho(t - \tau) e^{-i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} e^{i(\omega_L + \omega_{\text{US}})t}, \quad (2.9)$$

where (η, ξ, L) denotes the coordinates of the plane of the camera sensor. τ is the time delay introduced between the modulations of the reference beam and the ultrasound which allows selecting the depth of observation in the medium of study. The reference field is superimposed on the camera to the scattered field E from the turbid medium. E_c carries the contributions of all the photons out-coming from the sample, at frequencies ω_L for untagged ones, $E_{M\pm}$ at $\omega_L \pm \omega_{\text{US}}$ for linearly tagged ones. An image $I_\tau(\eta, \xi)$ is acquired during the integration time τ_c of the optical sensor:

$$I_\tau(\eta, \xi) = \int_{\tau_c} dt \left| E_0 \rho(t - \tau) e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} e^{i(\omega_L + \omega_{\text{US}})t} + E_c(\eta, \xi, L, t) e^{i\omega_L t} + E_{M+} e^{i(\omega_L + \omega_{\text{US}})t} + E_{M-} e^{i(\omega_L - \omega_{\text{US}})t} \right|^2. \quad (2.10)$$

Following the principle of the off-axis holography (see [subsection 1.6.3](#)), this recorded image is filtered in the spatial Fourier domain so as to only keep the tilted component. This operation leads to the following hologram, resulting from the interferences between the scattered light and the reference beam.

$$H_\tau(\eta, \xi) \propto e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} \int_{\tau_c} dt E_0^* \rho(t - \tau) E_{M+}(\eta, \xi, L, t). \quad (2.11)$$

The complex component around the off-axis lobes is proportional to the optical field of the acousto-optic tagged light. This interference signal gathers the contributions of all virtual source located on the y axis along the acousto-optic interaction area inside the medium. It writes as:

$$s(\tau) = \int_{\tau_c} dt \rho(t - \tau) \int_y dy \Gamma(y) \rho(t - c_s^{-1} y), \quad (2.12)$$

$$= \int_y dy \Gamma(y) M_\tau(y), \quad (2.13)$$

where $\Gamma(y)$ is our desired acousto-optic profile, the virtual sources, developed along the acoustic column and $M(y)$ is a tagging function.

$$M_\tau(y) = \int_{\tau_c} dt \rho(t - c_s^{-1} y) \rho(t - \tau). \quad (2.14)$$

Experimentally we adapt the length of the random pattern to the integration time of the camera $\tau_c = N T_0$ and we synchronize the beginning of the modulation width the triggering of the camera. For clarity of the demonstration we change the variables by

$t' = t - c_s^{-1} y$ and $u = \tau - c_s^{-1} y$. Hence u corresponds to the temporal mismatch between the two patterns at a given depth y on the acoustic column.

$$M_\tau(y) = \int_{-\frac{T_0}{2} - c_s^{-1} y}^{\frac{(2N-1)T_0}{2} - c_s^{-1} y} dt' \rho(t') \rho(t' - u). \quad (2.15)$$

By introducing Equation 2.6 in the Equation 2.14 we write:

$$M_\tau(y) = \int_{-\frac{T_0}{2} - c_s^{-1} y}^{\frac{(2N-1)T_0}{2} - c_s^{-1} y} dt \left[\sum_{m=0}^{N-1} b_m \Pi\left(t - \frac{m}{N} T_0\right) \right] \left[\sum_{n=0}^{N-1} b_n \Pi\left(t - u - \frac{n}{N} T_0\right) \right]. \quad (2.16)$$

Let us consider three cases:

- if $|u| < T_0/2N$, we have partial overlap (see Figure 2.7). We break the expression in two parts: one for the partial overlap of the two patterns, the second corresponding to anti-correlated portions:

$$M_\tau(y) = (T_0 - |u|) + \frac{|u|}{N} \sum_{k=0}^{N-1} \sum_{p=k}^N b_k b_p. \quad (2.17)$$

The second term is a sum of random equiprobable values of -1 and 1 . Its distribution is centered on 0 with $\sqrt{N}$ standard deviation.

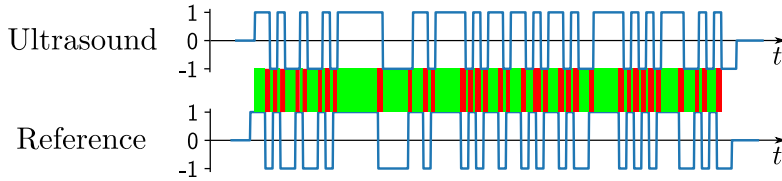


Figure 2.7 – Random modulation pattern modulating the ultrasound signal and the reference beam. When the delay τ between the patterns is null they interfere **constructively**. With a shift thinner than the duration of a jump the interference is successively **constructive** and **destructive**.

- else, if $T_0/2N \leq |u| < (2N-1)T_0/2$, the two patterns are totally uncorrelated. The signal consists in random fluctuations centered on 0 with $\sqrt{N}$ standard deviation (see Figure 2.8).
- finally for $T_0/2 \leq |u|$, the two patterns temporally do not overlap $M_\tau(y) = 0$.

Let us consider the triangle function:

$$\Lambda(y) = \begin{cases} 1 - |y| & \text{if } |y| < 1/2 \\ 0 & \text{otherwise} \end{cases}. \quad (2.18)$$

On first approximation if we neglect the $\sqrt{N}$ fluctuation on the sides, we consider $M(y)$ as a triangle function (see Figure 2.9):

$$M_\tau(y) = T_0 \Lambda\left(\frac{N(y - c_s \tau)}{c_s T_0}\right). \quad (2.19)$$

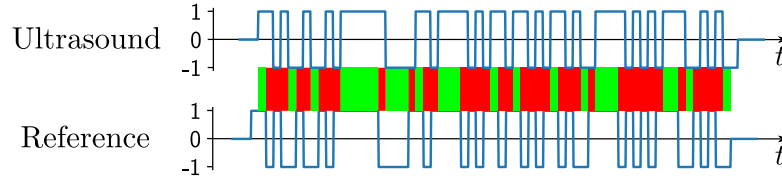


Figure 2.8 – Random modulation pattern made of 64 phase jumps modulating the ultrasound signal and the reference beam. When the delay between the patterns is greater or equals to the width of a phase shift their interference is successively **constructive** and **destructive**. As a results the overall contribution fluctuation is around $\pm\sqrt{N}$, N being the total number of phase jumps in the pattern.

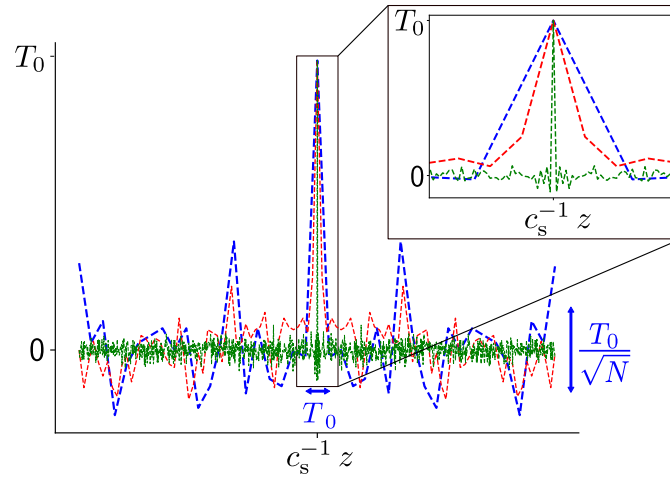


Figure 2.9 – Auto-correlation $M_T(y)$ of the random pattern for $N = 40, 100, 1000$ keeping the same pattern length.

According to [Equation 2.12](#), we have shown that modulating the ultrasound wave and the reference beam by a random-phase-jump pattern allows measuring a pointed signal. The width of the axial selection of the optical tagged field δy is given by the width of the jumps multiplied by the celerity of sound in the medium ([Lesaffre et al., 2009](#)):

$$\Delta y = \frac{c_s T_0}{N}. \quad (2.20)$$

As a result, if one wants a 1.5 mm-resolution, one chooses $T_0 = 1 \mu\text{s}$ as the width of a jump, which corresponds to 5 cycles per jumps for a 5 MHz carrier frequency. This method allows resolving with an accuracy identical to the pulsed one. Furthermore this method reveals a contrast proportional to the square root of the total number of jumps used and is more sensitive as it collects light along a longer period. The autocorrelation of the random patterns enhances the optical contrast at depth $y = c_s \tau$ by summing

the contribution of all the scattered light interacting with the ultrasounds during the integration time following a triangle law.

Chirp

Another use of chirped ultrasonic pulses consists in using the reference to produce autocorrelation on the camera chip during its exposure time. We based this section on the previous demonstration made for the random phase jumps. Chirp signal consists in a pulsed wave in which the frequency is linearly swept. Thus the phase of the ultrasonic wave writes as:

$$\phi_{\text{US}}(t) = 2\pi (\omega_{\text{US}} + b t) t \quad b = \frac{\Delta\omega}{T_0}, \quad (2.21)$$

with $\Delta\omega$ the amplitude of the sweep and T_0 the duration of the pattern, see Figure 2.10. The reference field is also swept in frequency on the same temporal length but delayed

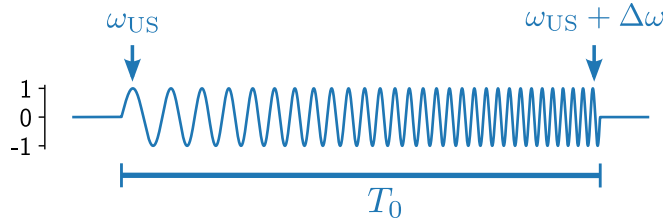


Figure 2.10 – Example of a chirp modulation. A pulse of temporal width T_0 is linearly swept in frequency such as the first cycle correspond to the pulsation ω_{US} and the last one matches the pulsation $\omega_{\text{US}} + \Delta\omega$.

of τ . Its phase thus writes as:

$$\phi_{\text{ref}}(t - \tau) = 2\pi [\omega_{\text{US}} + b(t - \tau)] (t - \tau). \quad (2.22)$$

The interferogram integrated by the camera carries the tagged-photon contribution. This signal, integrated on the integration time of the camera τ_c , writes as:

$$s(\tau) = \int_0^{\tau_c} dt e^{-i\phi_{\text{ref}}(t-\tau)} e^{i\phi_{\text{US}}(t)}, \quad (2.23)$$

$$= \int_0^{\tau_c} dt e^{2i\pi\{(\omega_{\text{US}}+bt)t - [\omega_{\text{US}}+b(t-\tau)](t-\tau)\}}, \quad (2.24)$$

$$= \int_0^{\tau_c} dt e^{2i\pi\tau(\omega_{\text{US}}-b\tau+2bt)}, \quad (2.25)$$

$$= e^{2i\pi\tau(\omega_{\text{US}}-b\tau)} \int_0^{\tau_c} dt e^{4i\pi b\tau t}, \quad (2.26)$$

$$= e^{2i\pi\tau(\omega_{\text{US}}-b\tau)} \frac{e^{2i\pi b\tau\tau_c}}{4i\pi b\tau} (e^{2i\pi b\tau\tau_c} - e^{-2i\pi b\tau\tau_c}), \quad (2.27)$$

$$= \tau_c e^{2i\pi\tau[\omega_{\text{US}}+b(\tau_c-\tau)]} \text{sinc}(2\pi b\tau\tau_c), \quad (2.28)$$

with $\text{sinc} : t \mapsto \sin(t)/t$ denotes the sinc function. As a result, the temporal autocorrelation of the two chirped modulation is a sinc function centered at $\tau = 0$. The width of the central peak $\Delta\tau$ is given by:

$$\Delta\tau = \frac{1}{b\tau_c} = \frac{T_0}{\Delta\omega\tau_c}. \quad (2.29)$$

Experimentally, the temporal length of the modulation is chosen equal to the integration time of the camera:

$$\Delta\tau = \Delta\omega^{-1}. \quad (2.30)$$

Thus, the accuracy of the selection is directly controlled by the frequency extension of the chirp $\Delta\omega$ (Figure 2.11). For example, a target resolution of 1.5 mm corresponds to a 1 MHz sweep in frequency. This autocorrelation corresponds to the tagging function,

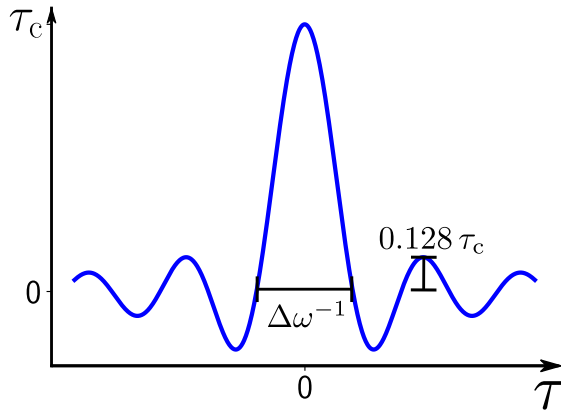


Figure 2.11 – Tagging function for a chirp modulation with a amplitude of the frequency-sweep $\Delta\omega$.

i.e. the point spread function of the virtual acousto-optic source. By tuning the delay τ of the reference beam, one can precisely control the position of central spike along the acoustic-axis $y = c_s \tau$.

In conclusion, for a given camera integration time τ_c random-phase-jump and chirp modulation provide a tight selection along the acoustic propagation axis. Nevertheless, the shapes of their associated point spread functions have an experimental incidence, as we will see in the next section.

2.1.5 Controlling the modulation using a rapid photodiode

Chirp signals provide a smoother modulation whereas random-phase-jumps has amplitude discontinuities. These discontinuities are manifested by a large spectral modulation of the carrier, inducing high-frequency components. Experimentally the emitters, such as the ultrasonic probe or the acousto-optic modulators (AOM), have a limited bandwidth. Because AOM and the US probe have not the same bandwidths, the modulations applied on the two arms of the holographic setup are not the same. Nevertheless, the autocorrelation of random-phase-jump is ‘naturally’ apodized by its random statistics

whereas the sinus-cardinal shape of chirp autocorrelation have lateral bounces around 0.128 robust to averaging. This means random phase patterns including more than 61 jumps² have a better contrast than chirp. In the chapter 3 we use $N = 2000$ long pattern which significantly overcomes this balance.

In order to monitor the phase modulation developed in the previous sections, we built a diagnosis arm on our experiment. This extension of the optical setup, presented on Figure 2.12, consists in a lens focusing the object and the reference beam on a photodiode and a camera. We use a tiny optical beam of few square millimeters that crosses the acoustic field emitted by a mono-element transducer. In a ballistic propagation regime, the photons are diffracted by acousto-optic effect along the propagation direction of the acoustic wave and thus can be easily discriminated using a simple spatial filter. We select the zero-order diffracted by the ultrasounds and focus it onto a fast photodiode and superimposed with a reference beam. A optical scheme is presented on Figure 2.12. To ensure the superimposition and the mode-matching of the two beams, we duplicate the beam by a beam-splitter and conjugate a small camera with the sensor of the photodiode. This way we can monitor the plane of the photodiode while tuning the mirrors.

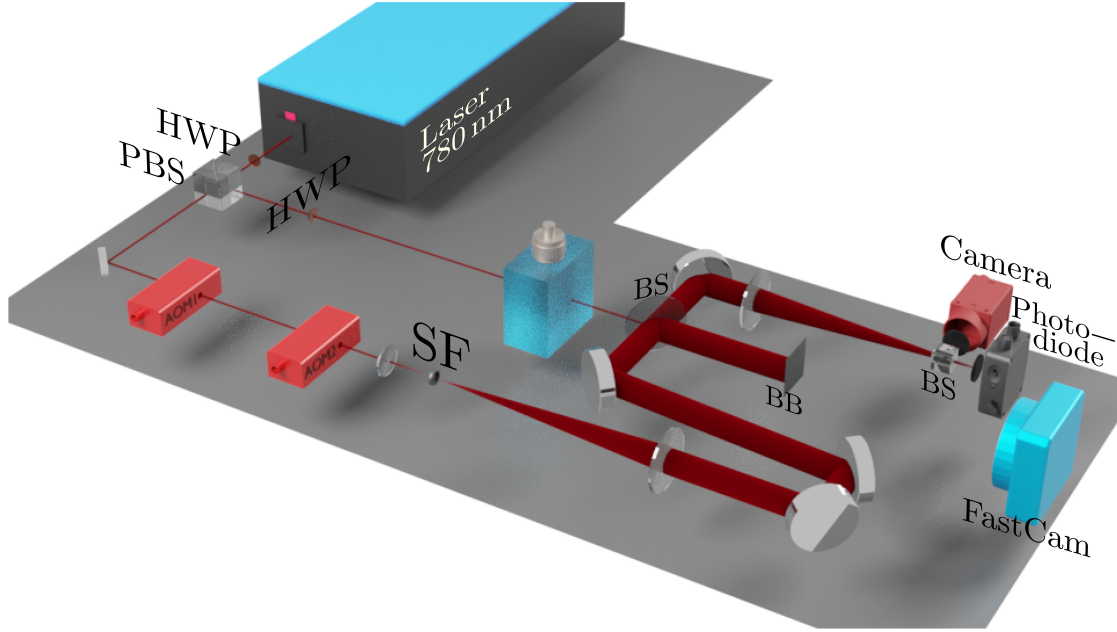


Figure 2.12 – Scheme of the phase diagnosis setup. At the output of the water tank in which the object beam and the modulated ultrasound interact, the beam is split to be focused by a lens to a photodiode and a camera. The sensors are protected from saturation by two power densities.

By recording the dynamic of the interference, we directly monitor the modulation.

2. As the contrast of random phase jumps PSF scales with $\sqrt{N}$. We need at least $N = 0.128^{-2} \approx 61$ jumps to overcome chirp-PSF contrast.

Figure 2.13a presents only the signal of the zero-order of the probe beam passing through the ultrasound focus. The acousto-optic tagging is manifested by a depletion of the zero-order as part of the light is diffracted out of the optical axis. Thus, Figure 2.13a shows a step in the signal received by the photodiode at the passing of the 200 μ s-random-phase-jump pattern.

We repeat the acquisitions for different widths of phase jump in order to test the fidelity of modulation. For this, we compared the modulation applied by the acoustic with the one of the acousto-optic modulation. We test jump widths from 0.2 μ s, which corresponds to a period of the acoustic carrier at 5 MHz, to 20 μ s, corresponding to the tenth of the total pattern. We also test frequency-sweep from 4 MHz to 0.05 MHz. These two scales correspond to the same range of resolution, from 0.3 mm to 30 mm. For all signals, the AOM produces a modulation that perfectly matches the theoretical one. This is explained by the large frequency range, 80(15) MHz, of the AOMs. On the contrary, the acoustic probe has a narrower frequency range. The data-sheet of the probe only provides the impulse response of the probe, not its emission diagram. However, we estimate it by recording the response a wide-range frequency-sweep. Indeed, wide-range frequency-sweep provides a direct link between time and frequency. Knowing the beginning of the modulation allows associating each amplitude to the corresponding frequency. The result is shown on Figure 2.13b. The depletion curve recorded for a such sweep fits the response of the probe.

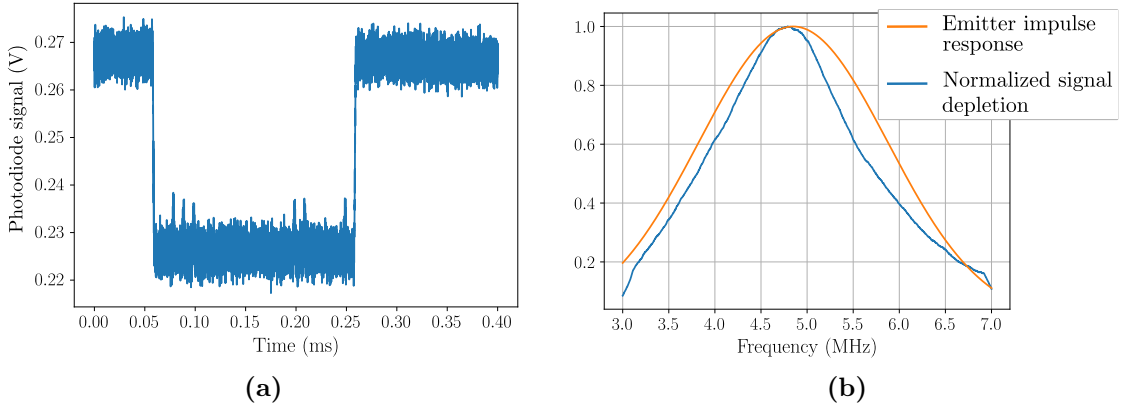


Figure 2.13 – Monitoring temporally-long modulations with a fast photodiode. (a) Photodiode signal depletion at the passing of a random-phase-jump modulation. (b) Normalized depletion signal at the passing of a 6 MHz wide-sweep modulation. Comparison with the impulse response diagram of the 5 MHz ultrasonic probe.

Moreover, all the signals that we tested are impacted by the spectral bandwidth of the probe. In order to limit this effect, we use 1 μ s-large jumps in the experiment presented in next chapters. Indeed, such pattern have spectral components spread over 1 MHz, in order to limit the impact of spectral bandwidth. In addition, this jump width allows putting 1000 jumps in a 1 ms-long pattern which corresponds to a contrast value

of 32 for the PSF.

2.2 Imaging using spatially focused ultrasonic beam

We proceeded to long-temporal modulation to scan the optical in high-scattering hydrogel. In this section, we present images acquired through an 2 cm-thick hydrogel with a reduced scattering coefficient of 10 cm^{-1} which corresponds to the typical scattering properties in human soft tissues (see [Table 1.1](#) on page 24). The turbidity of the hydrogel is given by the addition of Intralipid 10%. To create this controllable scattering we refers to the study of [van Staveren et al. \(1991\)](#) which describes the dosage necessary for a targeted reduced scattering coefficient in the 400 nm to 1100 nm spectral range. The dosage of agar is also adapted to reach a Young modulus of 50 kPascal, which corresponds to the mechanical properties of soft biological tissues. The recipe that we use follows the study of [Hall et al. \(1997\)](#). Two black-ink inclusions have been buried into the hydrogel to simulate an optical contrast.

We used the 5 MHz ultrasonic probe. As we have seen, it focuses at 50 mm, as the hydrogel is $4 \times 4 \times 2 \text{ cm}^3$ the probe could not be placed in contact with the sample. So we immersed it into a water tank to insure the impedance adaption.

The ultrasonic probe was fixed on a two-axis motorized translation stages to ensure displacements in the transverse-propagation plane. The third spatial degree of freedom, along the ultrasonic-propagation axis, was scanned by delaying the modulation applied on the reference beam, as see in [section 2.1.4](#). We modulate the reference and the ultrasonic wave by a pattern of 500 random phase jumps. Each jumps last $4 \mu\text{s}$ i.e. 12 cycles at 3 MHz which gives a total duration of 2 ms. The camera triggering is synchronized with the waveform generators of the modulation, see [Figure C.5](#) in the appendices for the scheme of electrical connection and [Figure C.6](#) for the chronogram of the signals. Its integration time of 2 ms fits the pattern duration. In these conditions, the selection produced by the random phase jumps method has a contrast $\sqrt{500} = 22$ and is 6 mm wide. Each point acquisition is averaged 100 times. We display the intensity of the acousto-optic signal, i.e. the square modulus of the Fourier components integrated around the off-axis lobes in the Fourier domain. Despite the 6 mm resolution, we observe the shadows of the two ink inclusions on [Figure 2.14](#). These shadows are smoothed by the large detection function and possibly by the diffusion of the black ink in the hours following the casting. The top-to-bottom intensity gradient of the acousto-signal explains by the attenuation of the ultrasounds in the sample ($-0.3 \text{ dB MHz}^{-1} \text{ cm}^{-1}$).

On [Figure 2.15](#), we present an acousto-optic image of light scattered across a turbid hydrogel, accompanied by the profiles drawn along the optical axis (top-right view) and across the input and the output planes (bottom-right view). This image is made of two acquisition stitched together on the optical axis. The discontinuity in the input surface profile is probably due to a little mismatch of coordinates as the intensity of the signal sharply varies close to the input-beam impinging area. With a transport mean free path $l^* = 1 \text{ mm}$, light enters in a multiple scattering propagation regime in the first half centimeter as we see with the axial profile decrease.

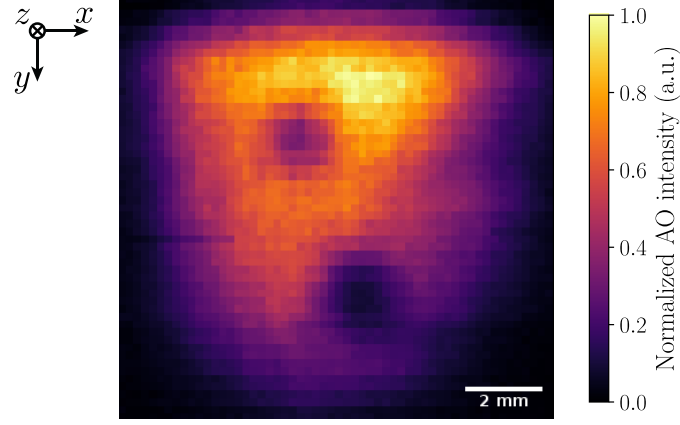


Figure 2.14 – Acousto-optic image of two ink inclusions buried in a 2 cm-thick scattering hydrogel, $\mu'_s = 10 \text{ cm}^{-1}$.

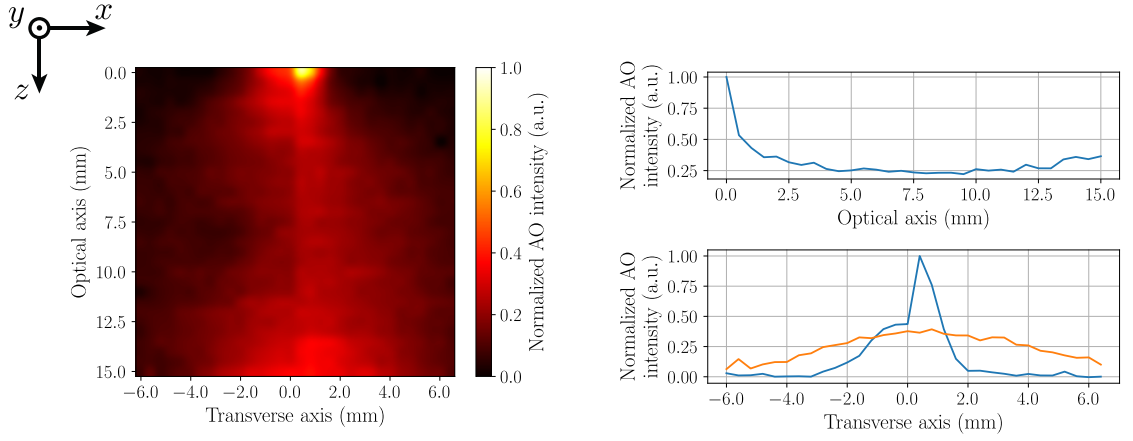


Figure 2.15 – Acousto-image of light scattering through a turbid hydrogel. Left: Acousto-optic image of the scattering of our 780 nm continuous laser across a turbid hydrogel. The hydrogel is 2 cm-thick with a reduced scattering coefficient of $\mu'_s = 10 \text{ cm}^{-1}$. The image was acquired by detecting the tagged-photon signal. We distinguish the shadow due to an ink-inclusion centered on position $(x, z) = (-2.5 \text{ mm}, 8 \text{ mm})$. Right: Profiles of the frequency shifted light intensity along the optical axis (top) and along the **input** and **output** faces (bottom).

Despite the fast frame rate of camera, 500 frames/second, the total acquisition time for these images raises up to $\sim 5 \text{ h}$. As the fast camera that we use does not provide frame streaming when triggered by an external signal, its internal memory need to be unloaded regularly along the measurement. The unloading demands around 25 minutes for each frame-batch.

2.3 The perspectives of non-linear acousto-optic tagging

The use of harmonic imaging in clinical context is well-known and common used in acoustic imaging (Tranquart et al., 1999). Indeed, ultrasonic pulses and other complex modulations such as random phase jumps and chirp patterns excite non-linear frequencies that broaden their spectrum by introducing replica of the modulation around ultrasound frequency multiple. Also, Selb et al. (2002) have already investigated the use of higher harmonics acousto-optic modulations for imaging. This approach gathers some advantages. First, as the non-linear generation of higher harmonic are local effects, one can emit a focused pulse at the fundamental frequency f_{US} and detect the light tagged by the non-linearly generated second harmonic at $2f_{\text{US}}$. Because of the frequency-dependency of ultrasound exponential attenuation ($-0.3 \text{ dB MHz}^{-1} \text{ cm}^{-1}$), it means that we reach deeper with high modulation frequencies. Secondly, as shown with simulations and experimentally by Selb et al. (2002), the point spread function of the tagging function at twice the ultrasound carrier ($2f_{\text{US}}$) is thinner by a $\sqrt{2}$ factor with respect to the fundamental point spread function. Nevertheless, the use of non-linear acousto-optic tagging demands high amplitude ultrasonic excitation which is not compatible with biomedical norms of 190 mW cm^{-2} for acoustic pulses (Phillips and Harris, 2008).

Another idea is to use long temporal-modulation such as random phase jumps combined with long camera integration to avoid sending high acoustic power into the sample. To form a $\{0, \pi\}$ random phase jump modulation at $2f_{\text{US}}$, we need to adapt the modulation to $\{0, \pi/2\}$ jumps. We made preliminary tests by measuring with an hydrophone the temporal pressure profile at the acoustic focus of the 5 MHz probe with standard $0-\pi$ pattern. As the arbitrary waveform generator that we use to generate random phase jumps cannot generate $\{0, \pi/2\}$ jumps. Indeed, the $\{0, \pi/2\}$ jumps have to be directly encoded with the phase of the carrier signal (which is impossible for a bandwidth limit reason) whereas $\{0, \pi\}$ jumps can be formed by multiplying together the carrier signal with the $\{1, -1\}$ modulation from two independent generators (one function generator for the carrier and one arbitrary waveform generator for the modulation). The solution is to play the modulation by an echographic device on which we can encode complex signals. We plan to implement this in the future. Combining on the same optical setup fundamental and harmonic detection using two specific reference arms at $f_{\text{L}} + f_{\text{US}}$ and $f_{\text{L}} + 2f_{\text{US}}$ would simultaneously provide two resolutions corresponding to the fundamental and harmonic point-spread functions.

2.4 Imaging using ultrasonic plane waves

In the previous sections, we have presented spatially-focused ultrasound. As we have seen, this focusing imposes to implement the scan of the ultrasonic probe, which requires time to explore the complete imaging volume. Then, the limited interaction area between light and sound provided by this method needs multiplying the acquisition to raise the SNR which also lengthens the imaging process. One of the main purposes of ultrasound-modulated optical tomography is to perform *in vivo* images. This demands to

shrink the acquisition time of coherent dependent signals shorter than the decorrelation time of living tissues, which is 1 ms. In order to maximize the tagging volume and maintain spatial resolution, new modalities of insonification based on multi-transducer probe were developed. [Laudereau et al. \(2016\)](#) demonstrates that the use of tilt ultrasonic plane waves can reconstruct the luminous intensity across a plane. This new type of insonification allows a drastic improvement of the imaging rate. This progress was made possible by replacing the former single-element probe by an ultrasound multi-element array. Indeed, the tagging volume is two orders of magnitude greater for plane wave imaging. With an array of transducer emitting a plane wave pulse, the interaction area scales at 5 MHz with:

$$\lambda_{\text{US}}^2 L = 0.3 \times 0.3 \times 40 \text{ mm}^3 = 3.6 \text{ mm}^3, \quad (2.31)$$

whereas with spatially-focused ultrasounds all the dimensions are given by the wavelength:

$$\lambda_{\text{US}}^3 = 2.7 \times 10^{-2} \text{ mm}^3. \quad (2.32)$$

The quantity of collected signal is significantly improved which allow dividing the averaging by a factor 50. As each of this successive emitters are independently controllable by a multichannel arbitrary waveform generator, the plane wave is tilted by an angle θ with respect to a delay law along the axis of the probe (see [Figure 2.16](#)). As the amplitude of the tilt is limited to $\pm 20^\circ$ by the transducer pitches, resolution in the axial direction is degraded compared to the isotropic one. [Laudereau et al. \(2016\)](#) combined two measurements with the ultrasonic probe at 90° to equilibrate the resolutions.

[Bocoum et al. \(2019\)](#) brought improvements on this technique by adding a spatial structuration. This consists in switching on or off some emitters on the echographic probe with respect to a spatial period λ_s (see [Figure 2.17](#)). This modulation adds more latitude to reconstruct horizontal Fourier components and allows reconstructing the image plane without moving the acoustic probe.

2.4.1 Fourier-transform acousto-optic imaging

Fourier-Transform Acousto-Optic Imaging (FT-AOI) was first introduced using spatially focused US by [Barjean et al. \(2015\)](#) and [Barjean et al. \(2016\)](#). There, a long US pulse is periodically modulated either in amplitude or phase. If the same modulation is reproduced on the reference arm, tagged photons appear as if generated by several points periodically distributed along the direction of propagation. One acquisition gives access to one component of the image Fourier Transform. In a recent article, [Bocoum et al. \(2020\)](#) have shown that using a transducer array, the same idea could be extended to fetch the two-dimension Fourier components of the image with a photorefractive-holography setup.

The purpose of FT-AOI is to broaden the tagging volume while keeping the resolution of the imaged plane. Similarly to the selection of light encoded by random phase jumps or chirped ultrasounds (see [subsection 2.1.4](#)) in FT-AOI the selection among tagged photons is performed during the integration time of camera. The following theoretical section analytically explicits the selection process.

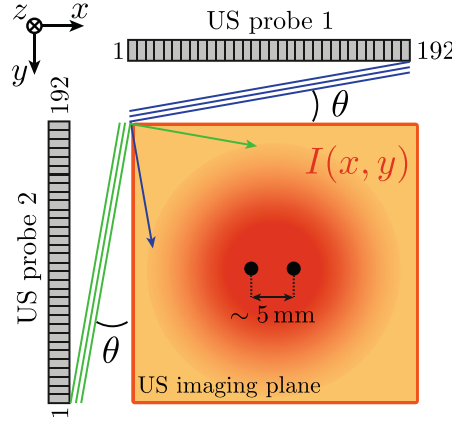


Figure 2.16 – Scheme of the tilted plane wave ultrasound-modulated optical tomography, adapted from the original scheme of [Laudereau et al. \(2016\)](#). A tilted plane-wave pulse is emitted by a 192-transducer array throughout the illuminated sample. The tilt angle θ is created by imposing a delay law along the array. $I(x, y)$ the scattered intensity profile in the ultrasound plane. In order to create an optical contrast, two ink inclusions, separated by 5 mm, are buried with the imaged sample. Two measurements, with the probe moved to a perpendicular face of the sample, allows reconstructing the imaged plane with the same resolution in the two dimensions.

2.4.2 Theoretical framework

The theoretical framework of FT-AOI is described by [Barjean et al. \(2016\)](#). In this study, an ultrasonic probe emits a temporally long pulse that is modulated by a binary periodic pattern, illustrated on [Figure 2.18](#) adapted from [Bocoum et al. \(2020\)](#). This one-dimensional excitation has then been extended to two dimensions using a multi-element probe by [Bocoum et al. \(2020\)](#).

The ultrasonic excitation

In this section, we develop the formalism for FT-AOI in two dimensions. For this, the pressure field is periodically structured along x and y . The x -axis refers to the direction along the transducer array and y to the acoustic propagation axes. The reconstruction of an image requires to perform multiple acquisitions. For each acquisition, the structuring frequencies are increasing harmonics that we index by m and n . In addition, by controlling the emission time, we can monitor the relative phase-shift ϕ of the structuring pattern. We note $P_{m,n}^\phi(x, t)$ the pressure wave emitted by the transducer array for the (m, n) structuring pattern. The pressure next to the probe, *i.e.* at $z = 0$ and for $0 \leq x \leq L$, where L is the width of the US probe, at $z = 0$ equals:

$$P_{m,n}^\phi(x, t) = P_0 h_{m,n}^\phi(t, x) \cos \omega_{UST}. \quad (2.33)$$

construction, we also impose that function $x \mapsto h_{m,n}^\phi(t, x)$ be L/m -periodic, where we recall that L denotes the length of the probe. Here below is the analytical expression of function $h_{m,n}^\phi$:

$$h_{m,n}^\phi(t, x) = \sum_{k=0}^{n-1} \Pi_n \left(t - \frac{m \nu_x^0}{n \nu_y^0} \frac{x}{c_s} - \frac{k T_0}{n} + \phi \right) \quad \Pi_n(t) = \begin{cases} 1 & \text{if } 0 \leq t < T_0/2n \\ 0 & \text{otherwise} \end{cases}. \quad (2.34)$$

Examples for this function are presented on [Figure 2.19](#). By neglecting diffraction effects,

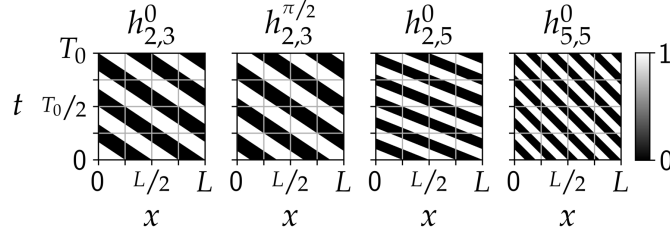


Figure 2.19 – Examples of the amplitude of modulation $h_{m,n}^\phi$.

we write the pressure wave within the medium at any depth y as:

$$P_{m,n}^\phi(x, y, t) \equiv A_{m,n}^\phi(x, y, t) \cos \left[\omega_{\text{US}}(t - c_s^{-1} y) \right], \quad (2.35)$$

$$A_{m,n}^\phi(x, y, t) = P_0 h_{m,n}^\phi(t - c_s^{-1} y, x), \quad (2.36)$$

with $c_s = 1540 \text{ m s}^{-1}$ is the velocity of sound across the sample and P_0 the nominal pressure. As a result, the optical field of the tagged photons in the US plane is modulated during the propagation of the spatial periodic mask $h_{m,n}^\phi$ (see [Figure 2.20](#)). Due to the acousto-optic effect, the tagged photons are frequency shifted from the optical carrier ω_L at $\omega_L \pm \omega_{\text{US}}$.

The detection

In ([Bocoum et al., 2020](#)), the tagged-photon signal is detected using two-wave mixing in a SPS:Te-photorefractive crystal. In this subsection, we develop the off-axis holographic detection.

In order to detect these photons, we modulate an auxiliary field at period T_0/n on a carrier frequency tuned in order to match tagged photons' one, *e.g.* $\omega_L + \omega_{\text{US}}$. The temporal expression of this reference field is set, on the surface $(\eta, \xi, z = L)$ of the camera sensor, to:

$$E_{\text{ref } n}(\eta, \xi, L, t) = E_0 \sin^2 \left(\pi \frac{n}{T_0} t \right) e^{-i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} e^{i(\omega_L + \omega_{\text{US}})t}, \quad (2.37)$$

where c is the speed of light, E_0 is the amplitude of the field, α_η and α_ξ correspond to the off-axis angles. The $\sin^2$ term corresponds to the T_0/n -periodic modulation, applied

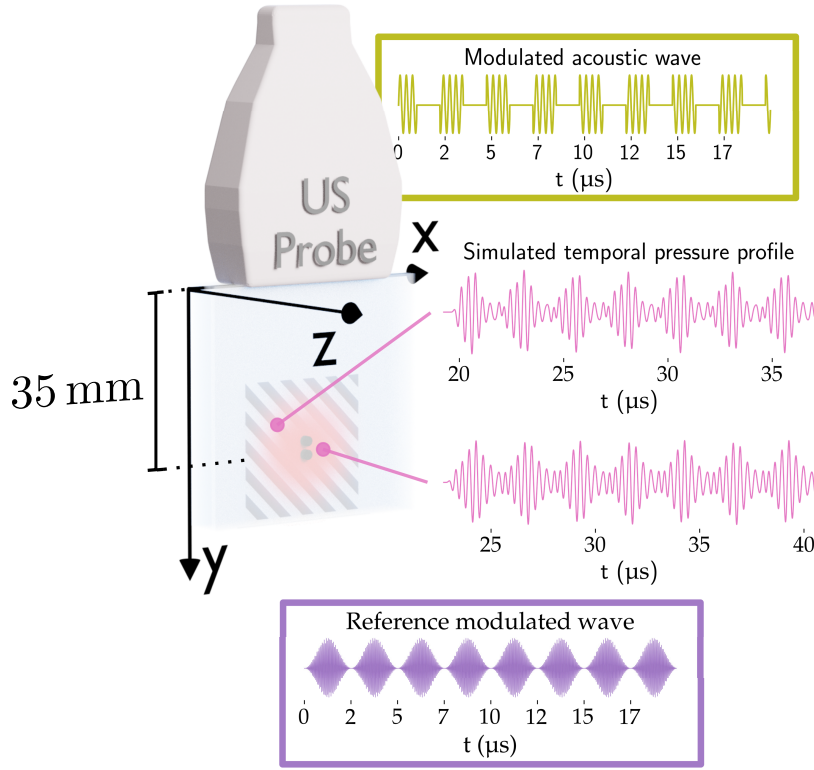


Figure 2.20 – Scheme of the optical probe beam modulation across the scattering sample by structured acoustic plane waves. The two ink inclusions to be imaged, represented in black at the center of the scattered light spot, are placed at the elevation focus of the probe, *i.e.* 35 mm. The drawn structure corresponds to $(m, n) = (3, 8)$ during its propagation accompanied with examples of two simulations of the acoustic wave temporal profile at different locations in the US plane.

together with the $h_{m,n}^\phi$ corresponding modulation. The reference field is superimposed on the camera to the scattered field E from the turbid medium. E hence carries contributions of all the photons out-coming from the sample, at ω_L for untagged ones, at $\omega_L \pm \omega_{US}$ for linearly tagged ones. An image $I_{m,n}^\phi(\eta, \xi)$ is acquired during the integration time of the optical sensor τ_c :

$$I_{m,n}^\phi(\eta, \xi) \propto \int_{\tau_c} dt \left| E_0 \sin^2 \left(\pi \frac{n}{T_0} t \right) e^{i(\omega_L + \omega_{US})t} e^{-i \frac{\omega_L + \omega_{US}}{c} (\alpha_\eta \eta + \alpha_\xi \xi + L)} + E^\phi(\eta, \xi, L, t) \right|^2, \quad (2.38)$$

Following the basic principle of the off-axis holography (see [subsection 1.6.3](#)), this recorded image is filtered in the Fourier domain so as to only keep the tilted component. This operation leads to the following hologram resulting from the interferences between the

scattered light and the reference beam:

$$H_{m,n}^\phi(\eta, \xi) \propto e^{i\frac{\omega_L + \omega_{US}}{c}(\alpha_\eta \eta + \alpha_\xi \xi + L)} \int_{\tau_c} dt E_0^* E^\phi(\eta, \xi, L, t) \sin^2\left(\pi \frac{n}{T_0} t\right) e^{-i(\omega_L + \omega_{US})t} \quad (2.39)$$

The averaged square modulus of this hologram, $s_{m,n}^\phi \equiv \langle |H_{m,n}^\phi|^2 \rangle$, is proportional to the average intensity of the tagged photons selectively detected in the US plane (Barjean et al., 2016). We write it in the following form:

$$s_{m,n}^\phi \propto \int_{x,z} dx dy I(x, y) \left[M_{m,n}^\phi(x, y) \right]^2. \quad (2.40)$$

where $I(x, z)$ is our desired acousto-optic image within the medium and $M_{m,n}^\phi(x, z)$ is a tagging function. This cross-correlation integral and the selection process which derives from it can be explained more visually as following. The low frequency modulation on the reference arm matches the temporal n -indexed modulation respectively applied on the ultrasonic signals played by the probe transducers. This modulation acts as a stroboscopic imaging during the camera integration time. As the modulations on both arms are synchronized, the acquisition selects photons coming for a given static position identified by the tagging function. The tagging function can be computed from the approximated form of the pressure wave given in the Equation 2.35 using with the following cross-correlation Barjean et al. (2016):

$$M_{m,n}^\phi(x, y) = \frac{1}{\tau_c} \int_{\tau_c} dt \sin^2\left(\pi \frac{n}{T_0} t\right) A(x, y, t), \quad (2.41)$$

$$= \frac{P_0}{2} \left[1 + \frac{1}{\pi} \cos\left(\frac{2\pi m}{L} x + \frac{2\pi n}{c_s T_0} y + \phi\right) \right]. \quad (2.42)$$

The complete development of this derivation is presented in the appendices, Appendix B. Measurements for various phase jumps, $\phi \in \{0, \pi/2, \pi, 3\pi/2\}$, give access to the real and the imaginary parts of the Fourier components $\tilde{I}$ of the acousto-optic image:

$$s_{m,n}^{3\pi/2} - s_{m,n}^{\pi/2} + i \left(s_{m,n}^\pi - s_{m,n}^0 \right) \propto \tilde{I} \left(m \nu_x^0, n \nu_y^0 \right), \quad (2.43)$$

where we note $\nu_x^0 = L^{-1}$ and $\nu_y^0 = (c_s T)^{-1}$. The illumination across the US plane $I(x, z)$ is thus simply reconstructed by performing an inverse Fourier Transform (iFT).

2.4.3 Simulation of the tagging

To reconstruct our object, we use the model previously described where diffraction of the US is completely neglected (Equation 2.35 and Equation 2.41). To verify that hypothesis, we have performed a simulation of the actual pressure emitted by the probe we used in our later described experiment. Simulations were performed using Field II open-source software as already described by Jensen (2004). The autocorrelation of the simulated pressure given by the left term of Equation 2.41 is shown on Figure 2.21 for $m = -3, 0$ and 3 , $n = 8$ and x from 20 to 50 mm. This elevation range is thus

centered around the focusing distance of individual transducers of the probe. We observe that these simulated tagging patterns are not distorted by diffraction, and fit well with theoretical patterns (right term of Equation 2.41). We observe on Figure 2.21 that distortion effects appear for $|m| \geq 6$. They manifest as phase shift around $z = 36$ mm for (6, 8) and $z = 26$ mm and 40 mm for (13, 8). If we look at the corresponding spatial Fourier domains also presented on Figure 2.21, this phase distortions exhibit as doubled spectral components. This is clearly visible for the patterns (6, 8) and (13, 8). Distortion on (6, 8) enhances a second-order spatial harmonics. Distortion on (13, 8) plays as a slow amplitude modulation that splits the spectral component in two points. These phase distortions invalidate our reconstruction hypothesis that each pattern plays as a single spectral contribution. For this reason, we validate our imaging principle with an object placed at the elevation focus of the probe and limit the harmonic content to 10.

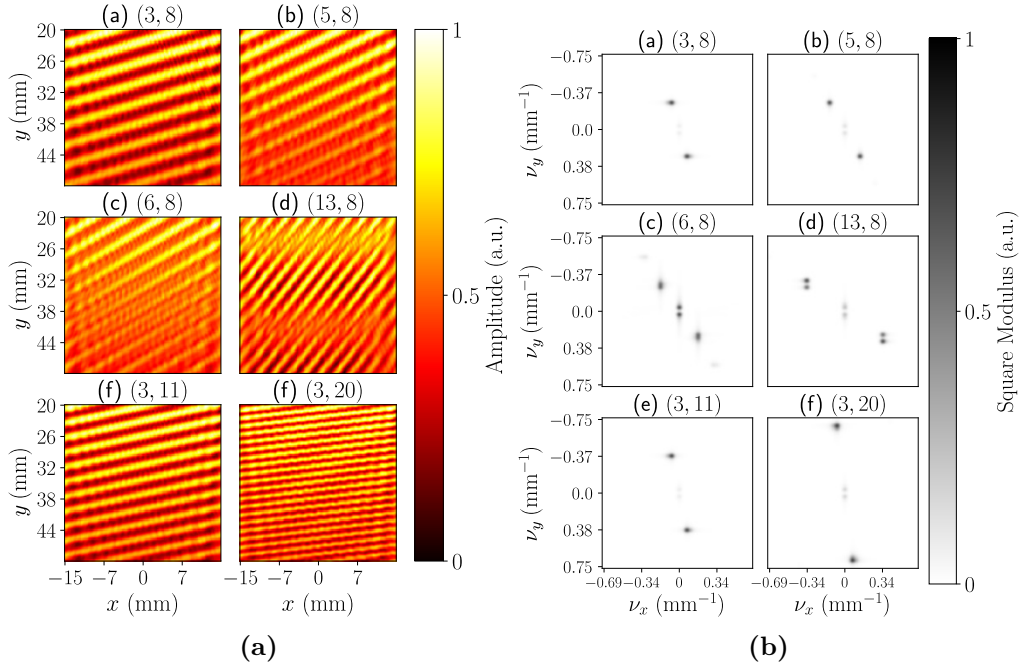


Figure 2.21 – Simulations of the tagging functions for 6 structures. Six couples of simulated tagging patterns, accompanied with their spectral components in the spatial Fourier domain, corresponding to the following (m, n) combinations (3, 8) (a), (5, 8) (b), (6, 8) (c), (13, 8) (d), (3, 11) (e) and (3, 20) (f).

2.4.4 The experimental setup

A preliminary work to demonstrate FT-AOI with off-axis holography was to remove possible sources of vibrations such as unstable pods or sources of temperature gradient surrounding the optical path. Then, as we have seen in the subsection 2.4.2, controlling

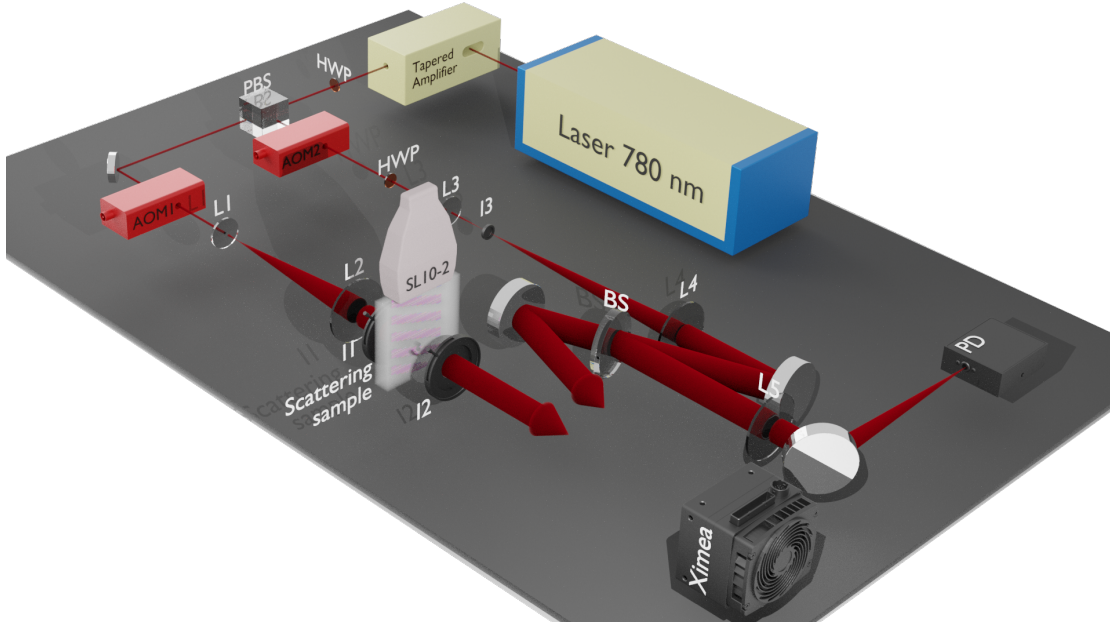


Figure 2.22 – Schematic of the optical setup used for FT-AOI. Light emitted by a laser diode at 780 nm is split between the probe beam and the reference arm with to a half-wave plate (HWP) followed by a polarizing beam-splitter (PBS). Both arms are then modulated in frequency with two acousto-optic modulators (AOM#) centered at 80 MHz with a relative difference of 3 MHz corresponding to the carrier frequency of the acoustic waves emitted by the transducer array (SL10-2 – Aixplorer SL10-2 US probe, SuperSonic Imagine, Aix-en-Provence, France) through the scattering hydrogel. Lenses (L#) and irises (I#) widen and filter the beams before they meet the camera (Ximea) sensor. The photodiode (PD) was used as a diagnosis tool for the modulation applied on the reference optical wave.

the phase of the modulation applied on the reference and the object arms is crucial for reconstructing an image. This imposes to precisely control the synchronization of each waveform generators employed in the experimental setup. A scheme presenting the electrical connections between all the involved devices and their chronogram are presented in the [section C.1](#). This connection scheme results from systematic study of all the possible desynchronizations. One of them was the impossibility to monitor the real instant and temporal length of the camera. Indeed, it appeared that the integration time of the camera chosen through the Application Programming Interface (API) of the camera does not correspond to the actual one executed by the camera³. For this

3. The other camera (Photron FastCam SA4) used for measuring the AOTM (see [chapter 4](#)) has a programmable output channel to monitor the actual integration signal

purpose, we added a diagnosis arm, picking up part of light of the reference beam and focus it on a fast photodiode (denoted PD on [Figure 2.22](#)). This diagnosis arm allowed us comparing the signal acquired by the camera with a simulated one reconstructed by integrating the temporal modulated intensity continuously recorded by the photodiode. With this, we tuned the triggering timing applied to the waveform generators and the camera. In order to overcome the uncertainty on the exact camera integration time, we extended it and gated the modulation applied to the generators.

The sample is a hydrogel mimicking both the optical and mechanical properties of soft biological tissues ([Jacques, 2013](#)). The total dimensions of the hydrogel along x , y and z axes are $5 \times 5 \times 1 \text{ cm}^3$. The mechanical properties is determined by the concentration of Agar, 2% which gives a Young modulus of 50 kPa. The concentration of Intralipid 10% is adjusted to obtain a reduced scattering coefficient $\mu'_s = 1 \text{ cm}^{-1}$ (transport mean free path $l^* = 1 \text{ cm}$). Agar is diluted into hot water at 90°C and Intralipid is added once the mixture has cooled down to 70°C . Two ink inclusions are inserted at a 5 mm depth. The dimensions of their cylindrical shapes are 4 mm in height and a diameter of 2 mm. Their centers are separated by 4 mm. A photograph showing the two inclusions taken during the molding is shown on [Figure 2.28](#). The sample is illuminated with a single-longitudinal mode laser centered at 780 nm. Directly at the output of the laser (MOPA, Sacher Lasertechnik GmbH), the incident power is about 370 mW for a beam diameter of 13 mm, corresponding to a section of 1.26 cm^2 . The transmitted-photon flux recorded by the camera (Ximea xiB-64) located at 34 cm from the sample output surface is $3.6 \mu\text{W}$. This distance matches the trade-off between the interference fringe sampling and the collection of tagged-photons. [Figure 2.23](#) shows the elevation of signal in the off-axis area in the 2D Fourier domain when the US are turned on. We only use a 1024×1024 pixels area of the 16-megapixel chip, to limit the processing

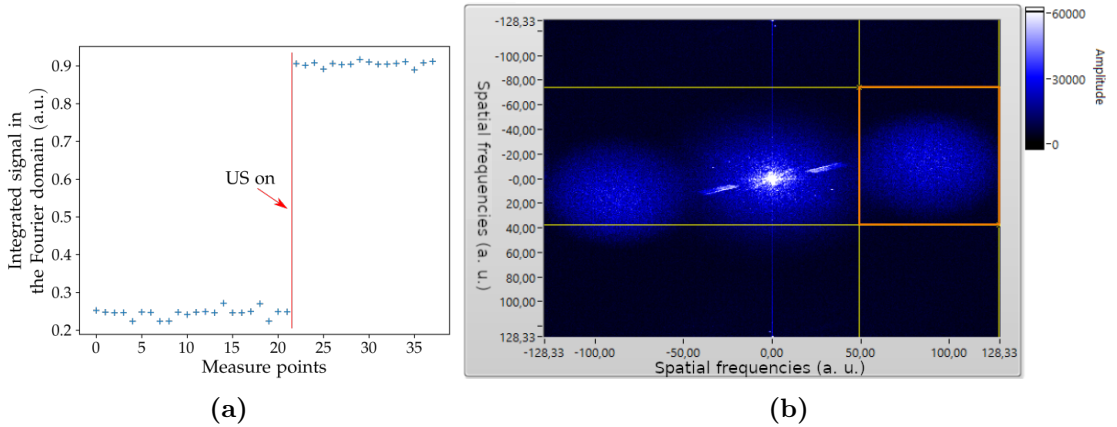


Figure 2.23 – For choosing the optimal output surface of the hydrogel to the sensor of the camera we studied the level of the AO signal in the Fourier domain. (a) presents the gap of signal level in the orange framed zone on (b) when the US probe emits. Screenshot of [Bocoum et al. \(2019\)](#)’s Labview graphical interface developed to control the FT-AOI setup.

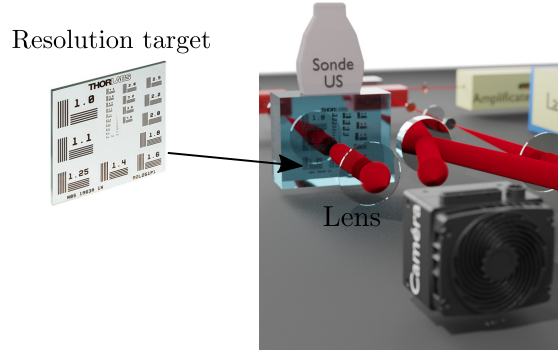


Figure 2.24 – For the ballistic validation experiment, the scattering sample is replaced by a resolution target immersed in a water tank. The US propagation plane and the camera sensor are conjugated through a lens.

time of raw images. A full frame acquisition (under progress) improves the SNR as the sensor surface is increased but would need further software optimization, for instance with the implementation of Graphics Processing Unit (GPU) for the reconstruction processing. The structured US plane waves are emitted with a commercial US probe (Aixplorer ultrasound imaging device SL10-2 US probe, SuperSonic Imagine, Aix-en-Provence, France). It consists in a linear array of 192 individual transducers (transducer pitch $200\text{ }\mu\text{m}$ for a total probe width of 3.8 cm) in the x -direction with a fixed focal length of 35 mm . With an excitation voltage of 15 V for the piezo-elements, the maximal pressure of the US wave was measured to be $\approx 300\text{ kPa}$. Such a pressure corresponds to a US peak intensity of $\approx 3\text{ W cm}^{-2}$. The US sequence pattern $P_{m,n}^\phi$, Equation 2.35, of $T_0 = 20\text{ }\mu\text{s}$ is repeated five times to match the exposure time of the camera $\tau_c = 100\text{ }\mu\text{s}$. The Aixplorer ultrasound device, which controls the US probe emission, is triggered together with the camera. According to the cyclic ratio of the US emissions, the pressure field corresponds to an average intensity of around 150 mW cm^{-2} within the medium, far below biomedical norms of 720 mW cm^{-2} (Phillips and Harris, 2008). For this acquisition the camera is triggered at 300 Hz with an integration time of $100\text{ }\mu\text{s}$ and each component is measured with a single-shot image. The total acquisition time to image a complete field of view of $30 \times 30\text{ mm}^2$ is 1.5 s .

2.4.5 Validation in the ballistic regime

A straight way to visually catch the principle of the Fourier transform imaging is to process first in ballistic regime. For this, we put aside for a moment our scattering hydrogel sample to replace it with a resolution target immersed into a water tank. The water is used to convey the US. The US propagation plane, tangent to the resolution target is conjugated with the plane of the camera sensor through a lens (see Figure 2.24).

This step allows controlling the proper selection of tagged photons at the US propagation plane. Indeed, according to the principle of off-axis holography, in a ballistic

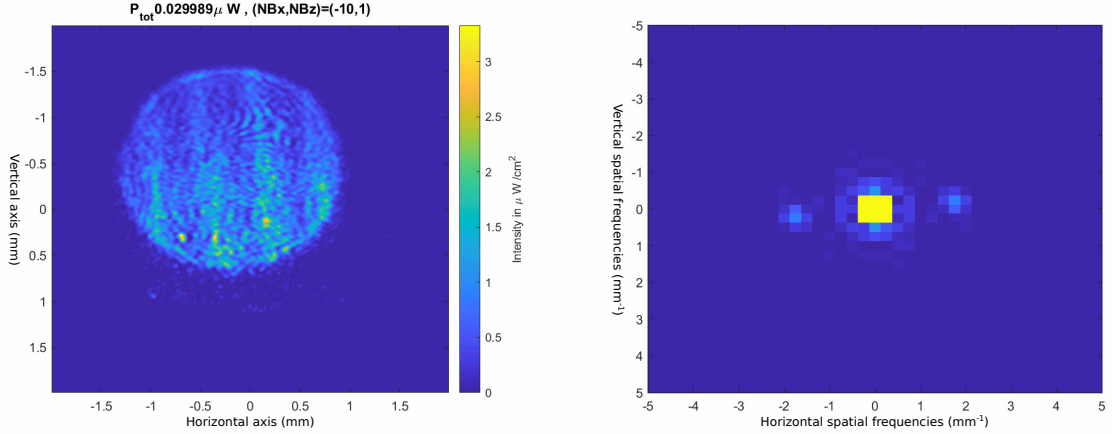


Figure 2.25 – The selection of tagged photons in the US propagation plane is directly reconstructed through ballistic imaging. **Left**-side image shows the tagged light intensity in the US propagation plane. It is reconstructed using off-axis holography. **Right**-side image shows the intensity in the spatial Fourier domain of the left-side image. The probed Fourier components are clearly distinguishable from either side of the central zero-order.

configuration, we directly reconstruct intensity map of the tagged light in the US propagation plane as shown on the left side of [Figure 2.25](#). The right side image shows the square modulus of its spatial Fourier domain. On this example one clearly identifies the inclination of our periodic pattern on the direct image and the corresponding two symmetrical spots on the Fourier spectrum. When we repeat the acquisitions for multiple Fourier components we reconstruct an image of the optical contrast. [Figure 2.26](#) shows the reconstructed images for two sets of Fourier components. If the halo of scattered light visible on the center of both images is correctly captured by low harmonics, thinnest structures of the bars are revealed for the right image with a vertical harmonic content from $n = 1$ to 10.

2.4.6 Imaging test on phantoms

We image the two ink inclusions hidden within hydrogels of two different forces of disorder. The image corresponding to the $\mu'_s = 1 \text{ cm}^{-1}$ hydrogel featured on [Figure 2.27](#) shows two dark spots in the halo of the tagged beam. The size of the inclusions, diameter 2 mm, and the gap between them, centers separated by 4 mm, corresponds to what we can measure on the photograph of the hydrogel, taken above the gel cast (see [Figure 2.28](#)). This image was reconstructed from the 110 complex Fourier components, $m \in \llbracket -5, 5 \rrbracket$, $n \in \llbracket 1, 10 \rrbracket$. The resolution of FT-AOI is given by the spatial period of the higher harmonics used in the structuring pattern $h_{m,n}$ with the wavelength of the US carrier as the ultimate limit. As the maximum values for m and n are respectively $m = 5$

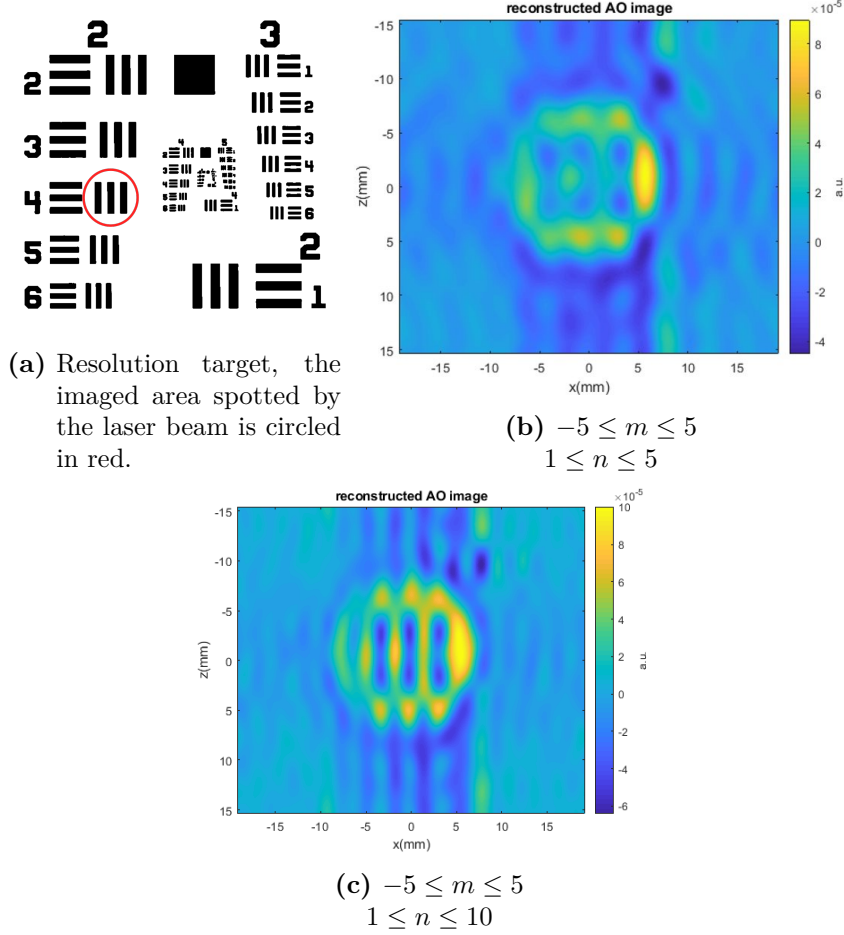


Figure 2.26 – FT-AOI reconstructed images of a resolution target (c) in ballistic regime. (b) is reconstructed from 55 measured Fourier components. (c) is reconstructed from 110 measured Fourier components. We distinguish the light disk with three vertical shadows corresponding to the three 1.6 mm-width vertical bars circled in red on (a). The increase of contributing components highlights the profiles of the bars.

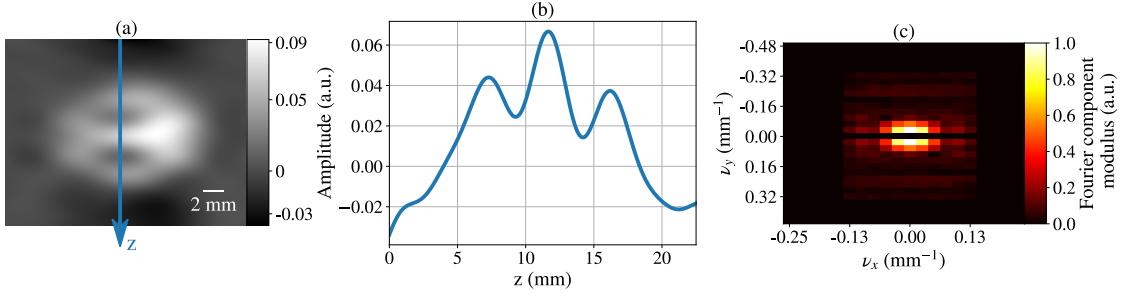


Figure 2.27 – FT-AOI of two ink inclusions buried within a 1 cm-thick scattering hydrogel with a reduced scattering coefficient of 1 cm^{-1} . (a) The image is obtained by gathering the 110 experimentally measured Fourier components for $-5 \leq m \leq 5$ and $1 \leq n \leq 10$. The profile (b) is drawn following the blue line on the acousto-optic image crossing the two dark spots. (c) Modulus of the 4-phase acquired Fourier components.

and $n = 10$, the resolutions along x and y are respectively $(m \nu_x^0)^{-1} = 7.7 \text{ mm}$ and $(n \nu_y^0)^{-1} = 3 \text{ mm}$, which discriminates our two inclusions.

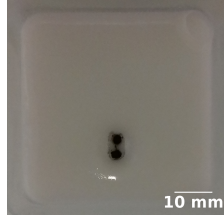


Figure 2.28 – Photograph of the two ink inclusions taken above the gel cast. The two inclusions stand at 35 mm from the top surface of the gel. This distance corresponds to the elevation focus of the probe.

In order to illustrate the greater time-efficiency of FT-AOI, we draw a comparison with AO imaging in real space as for instance performed using spatially-focused US with random phase jumps modulation such as in the [subsection 2.1.4](#). In this respect, let us assume such an image be sampled every $\sim 2 \text{ mm}$ over a $30 \times 30 \text{ mm}^2$ field of view. This corresponds to $15 \times 15 = 225$ points to acquire. For each sampled point, the AO detected signal in experimental conditions similar as in present work is nearly one to two orders of magnitude lower than for FT-AOI. In fact, because the tagging volume is reduced to few cubic millimeters, it requires a proportional increase in the number of averaging. As a result, the acquisition time for an image acquired in real space is on the order of a minute. In FT-AOI where acquisition is performed in reciprocal space, the tagging volume is significantly increased such that averaging is no longer required. As a result, the method is typically one order of magnitude more time efficient.

For a stronger-diffusive hydrogel $\mu_s = 10 \text{ cm}^{-1}$, the transmitted intensity through the sample is measured 6 times weaker. This decreases the SNR to ≈ 1.6 still with single

shot measurements. The resulting noise upon the Fourier components, for a single-shot measurement, blurred the reconstructed image (see Figure 2.29). The two inclusions are still discriminated but the reconstruction of the illumination around is perturbed by low-amplitude high-spatial-harmonic components. We nevertheless emphasize that this result is obtained without averaging, thereby demonstrating the robustness of our imaging strategy.

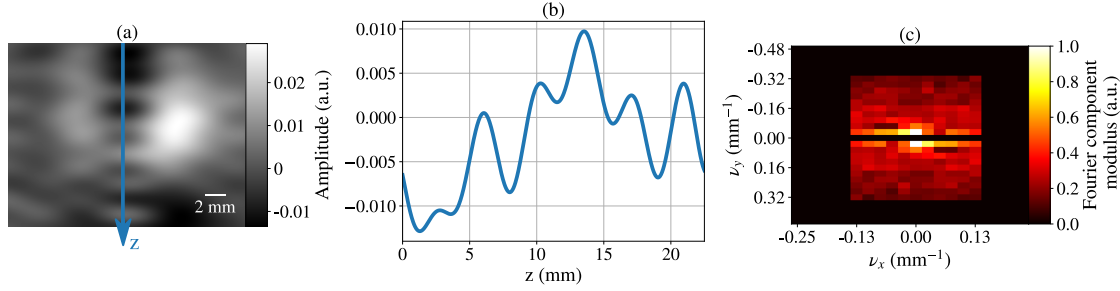


Figure 2.29 – FT-AOI images of two ink inclusions casted in a 1 cm-thick scattering hydrogel with a reduced scattering coefficient of 10 cm^{-1} . (a) The image is obtained by gathering the 110 experimentally measured Fourier components for $-5 \leq m \leq 5$ and $1 \leq n \leq 10$. The profile (b) is drawn following the blue line on the acousto-optic image crossing the two dark spots. (c) Modulus of the 4-phase acquired Fourier components.

The autocorrelation between scattered light from the sample and the reference beam is thus physically recorded during the integration time of the camera on multiple speckle grains. These larger spatial and temporal integrations allows collecting enough signal to perform a full acquisition without any averaging. In this condition, the imaging is robust against the fast decorrelation of living tissues. Furthermore, the use of US plane wave and long duration pulse limit the peak-power delivered into the sample to stay below biomedical norms. Finally, the overall acquisition time of 1.5 s, making this method adapted for *in vivo* monitoring. This work has been published in (Dutheil et al., 2021).

2.5 Conclusion and perspectives

In this chapter, we have presented the different modulation strategies that have been developed for acousto-optic imaging. We have seen that the use of echographic probe provides broad acoustic excitation allowing imaging a plane in an overall acquisition time compatible with *in vivo* prerequisites. FT-AOI is promising for future investigation in living tissues. Moreover improvement are foreseeable in the reconstruction of the images. Indeed we have seen with simulations, some ultrasonic patterns probe multiple Fourier components of the imaged plane. At this moment, this is a limitation for the diversity of patterns usable in FT-AOI. Nevertheless, we take advantage of these diffraction effects by measuring the tagging functions for each pattern in a known environment e.g. ballistic observation in a water tank. Then we could adapt the reconstruction taking into account

all two-dimensional Fourier spectrum of the tagging pattern including the effects of pattern edges and acoustic diffraction. To do so, we simulate the tagging function in the ultrasonic plane for each structured pattern. It would allow us to build a matrix of the spatial-spectral components of each structuring pattern in the spatial Fourier domain. Inverting the matrix would improve filling the Fourier components of the imaged plane. Then, using the matrix analysis operations that we will see in the following chapter we could engineer a basis patterns based on the principal component of such an acousto-optic tagging matrix.

CHAPTER 3

Focusing light in scattering media using the transmission matrix

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As we have seen in the previous chapters, the acousto-optic imaging has been significantly improved by the use of transducer arrays. The transducers array used for structured plane wave and Fourier-transform imaging includes 192 independently controllable transducers. The multiplication of emitters thus offers a new degree of freedom and new possibilities to sample the optical contrast within the medium. On the optical side, the development of digital devices to modulate the optical wavefront allows probing the transmission and reflection channels of a scattering sample. In this section, we will show how optical wavefront shaping techniques can be used to measure the optical transmission matrix of scattering media.

3.1 Optical wavefront control

Optical-phase correction devices were first developed in astronomy to compensate for atmospheric distortion of light. In the middle of the twentieth century, [Babcock \(1953\)](#) proposed such an optical setup based on deformable actuators and a wavefront sensor. Nowadays, this technique is implemented on most of telescopes. The topic is commonly named *adaptive optics*. These techniques were then transposed in microscopy applications, in particular for ocular imaging to compensate for ocular aberrations ([Zommer et al., 2006](#); [Jian et al., 2013](#)).

Nevertheless, adaptive optics reaches its limit for the strong aberration regime as the number of modes to be controlled exceeds the capabilities of deformable mirrors — typically up to few tens of mirrors. In the past fifteen years, wavefront shaping techniques have turned up as a tool of choice to control the propagation of light through scattering media, in particular for imaging applications ([Popoff et al., 2010b](#); [Xu et al., 2011](#); [Mosk et al., 2012](#); [Judkewitz et al., 2013](#); [Katz et al., 2014](#); [de Aguiar et al., 2016](#)). Wavefront shaping consists in spatially modulating the amplitude and/or the phase of the incident beam on a given propagation medium. Here we present the three most commonly used spatial light modulators.

3.1.1 Liquid crystal on silicon spatial light modulator (LCOS-SLM)

This technology is based on birefringence properties of liquid crystal ([Savage, 2009](#)). This device is presented as a reflective surface composed of a linearly-aligned nematic-liquid-crystal layer under a transparent electrode array (see the schematic in [Figure 3.1](#)). Liquids crystals consist in a nematic phase of organic rod-shaped molecules. When they are all oriented in the same direction, the liquid-crystal layer presents birefringence properties similar to some crystals with an ordinary and extraordinary optical indices. As a liquid phase, the global orientation of the molecules can tilt, thus changing the refractive index seen by light polarized parallel to the liquid crystal molecules. This orientation is sensitive to electrical tension, which allows controlling the refractive index of the liquid-crystal layer by applying a voltage through it ([Lueder, 2010](#)). Therefore, liquid-crystal cells are made of a liquid-crystal layer included between two electrodes driving the orientation of the molecules. As a result, LCOS-SLM provides a phase modulation from

0 to 2π at a given wavelength with around one million degrees of freedom, without any change in the light intensity and polarization direction. Nevertheless, the response time of liquid-crystal cells ($\sim 20 \mu\text{s}$) limits its speed to a few tens of hertz. In this thesis, we use a phase-only LCOS-SLM in reflection.

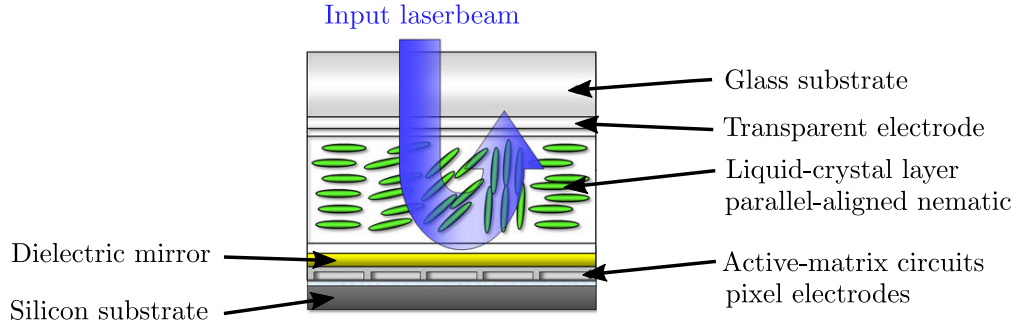


Figure 3.1 – Sectional drawing of a reflection liquid-crystal-SLM display, schematic adapted from the Hamamatsu’s device data-sheet. We see the liquid crystal layer sandwiched between two electrodes, the upper one is transparent, which control the molecule orientation within the nematic phase. The reflection is provided by a dielectric mirror under the liquid-crystal layer.

3.1.2 Micro-Electro-Mechanical-System-SLM (MEMS-SLM)

MEMS-SLM are similar to adaptive optical mirrors, as shown in Figure 3.2. They consists in an array of $10 \mu\text{m}$ to $100 \mu\text{m}$ flat mirrors mounted on individual linear actuators controlled using electrostatic forces. As a result the elevation of each mirror induces a modulation from 0 to at least 2π at a given wavelength. The low mechanical response of the actuators, $10 \mu\text{s}$ to $100 \mu\text{s}$, allows this technology to reach a 10 kHz to 100 kHz refresh rate.

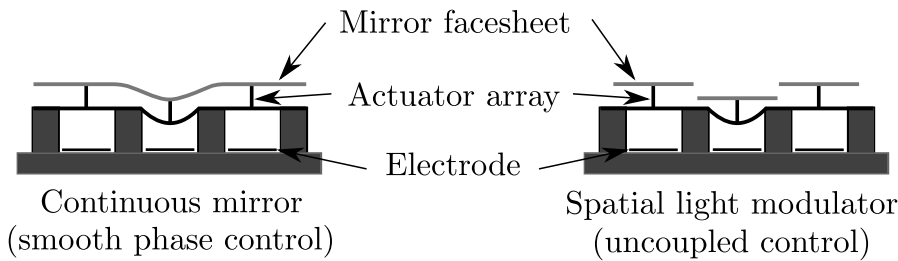


Figure 3.2 – Schematic of a micro-electro-mechanical-system-SLM. The left-side schematic shows a continuous surface mirror actuated by an array of electrodes. The right-side schematic shows a discrete array of mirrors individually controlled by actuators, providing an uncoupled control.

3.1.3 Digital Micro-mirror Devices (DMD)

DMDs are composed of tiltable $10\text{ }\mu\text{m}$ to $100\text{ }\mu\text{m}$ -pitch mirrors, as shown in [Figure 3.3](#). Each of them can independently and controllably flip between two angles when a voltage is applied. They provide a binary amplitude modulation in a direction of space. This technology can reach a refresh rate of a few tens of kilohertz. As explained by [Bifano and Stewart \(2005\)](#), this speed limitation of the actuator around one hundred of kilohertz is due to the mechanical constraint imposed by the viscosity of the surrounding air. Higher frequencies could be reached in vacuum enclosed devices. [Goorden et al. \(2014\)](#) demonstrated that a simple optical system based on DMD, lenses and spatial filters provides a spatial amplitude and phase modulation at the cost of a loss of resolution.

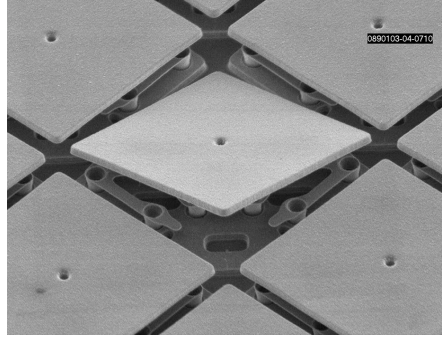


Figure 3.3 – Schematic of a digital micro-mirror device. The mirror at the center of the image is tilted at an angle opposite to its neighbors.

The [Table 3.1](#) summarizes the main characteristics of these commonly used SLMs.

Technology	LCOS-SLM	MEMS	DMD
Number of pixels	10^6	10^3	10^5
Refresh Rate (Hz)	$10^{1\cdots 2}$	10^5	$10^{4\cdots 5}$
Modulation	Full-phase $0 \cdots 2\pi$	Full-phase $0 \cdots 2\pi$	Binary amplitude on/off
Price (€)	10^4	10^5	10^3

Table 3.1 – Comparative table of optical wavefront control technologies.

The development of these technologies has lead to advances in numerous fields of applications. As we see in the following, these wavefront-shaping devices offer the possibility of measuring the transmission matrix of an optical system by sequentially modulating the input illumination throughout a basis. [Matthès et al. \(2019\)](#) study the propagation modes through a ground-glass diffuser and multi-mode fibers using a DMD. They also allow temporal shaping of optical signals. As an example, [Monmayrant et al. \(2010\)](#) explains temporal-shaping techniques, including the use of a grating mirror to diffract an optical beam on a SLM, thus providing a spectral dependency of the modulation. In

this work, we use a phase-only LCOS-SLM to produce a spatial light modulation. In the next section, we present the first experiment demonstrating light focusing through a scattering medium.

3.1.4 Focusing light through a complex medium

In a seminal experiment (Vellekoop and Mosk, 2007), Vellekoop et al. (2010) have managed to focus light through a scattering media using wavefront shaping. This experiment consists in iterating on the phase map of an input optical wavefront in order to overcome the effect of scattering to concentrate light in a tight spot (see Figure 3.4). In this experiment the SLM is divided into N individual segments. Starting from a flat-phase plane, the phase value of each segment is shifted from 0 to 2π . The phase corresponding to the brighter intensity in the target zone is kept (see Figure 3.5). The optimization then switches to the next segment. The overall enhancement factor η of this method is defined as the ratio between the optimized intensity and the initial average intensity and scales with the number of degrees of freedom N , i.e. the number of segments on the SLM (Vellekoop et al., 2010):

$$\eta_{\text{eff}} = \frac{\pi}{4}(N - 1) + 1. \quad (3.1)$$

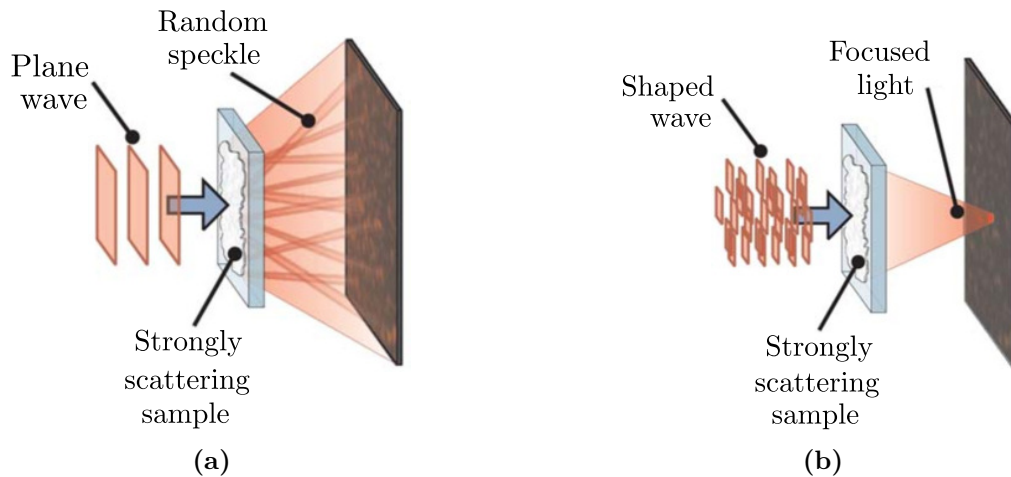


Figure 3.4 – Iterative focus optimization through a strongly scattering sample. Vellekoop and Mosk (2007)’s experiment schematic for focusing through a strongly scattering sample by sequentially wavefront shaping the optical wave incident to the sample. (a) At the initial step, the optical sensor records a complex speckle illumination. (b) At the final step, the process has adapted the input wavefront using the SLM to orient constructively the interference in a target region.

Indeed, focusing through scattering media offers new possibilities for imaging through or in complex media, develop *in situ* optical tweezers for biology protocols for example. In

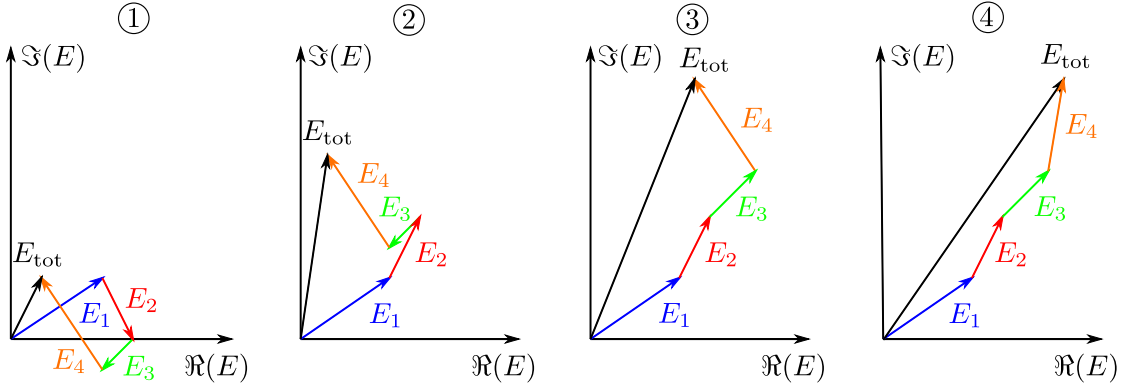


Figure 3.5 – Schematic of the principle of focusing on a target area in the $\mathbb{C}$ plane through sequential optimization. These graphs show the amplitude of the optical field in the target area in the $\mathbb{C}$ plane at the four different steps. The total optical field is the result of the summation of the contributions (E_i) originating from the different pixels. Initially, all the pixel contributions sum randomly. Throughout the optimization, the different contribution are rotated in a way that maximizes the modulus of the optical field in the target area. Outside the target area SLM pixels contribute randomly.

the next section, we explain how such optimizations are possible in volumetric scattering media with no direct access to the target plane.

3.2 Focusing light inside a scattering medium

Such technique based on a feedback optimization has proven to be efficient to focus through a scattering medium. However, it requires having access to both sides of the medium. The principal difficulty when targeting to focus into a scattering medium is that we cannot directly access to the target zone. For that reason, the methods that have been developed throughout the last decade implies so-called a guide-star component dipped inside the medium, interacting specifically with the illumination. To be used, the result of this interaction needs to be detectable from outside the medium in order to implement a feedback control.

The name guide-star originates from adaptive optics in astronomy when [Fugate et al. \(1991\)](#) used a laser to create a tight spot in the atmosphere. This artificial optical beacons serves as a target object of known geometry to adapt the surface of the deformable mirror to compensate for atmospheric distortions. The concept of this technique is to use a target object of known properties (shape, phase, absorption) embedded in the medium of study. The observation of the interaction between this target and the sent optical wavefront reveals information about the propagation properties between the input plane and the target.

Many teams use guide-star methods relying on different types of targets. [Katz et al. \(2011\)](#) use the enhancement of two-photon fluorescence to focus light behind a scattering layer. However, this implies introducing fluorescent markers in the sample, which is invasive. Therefore, non-invasive focusing have been demonstrated exploiting optical non-linearities. Scanning in depth is then done thanks to the memory effect ([Katz et al., 2014](#); [Tang et al., 2012](#)). In this section, we will present non-invasive guide-star-based focusing using the acousto-optic interaction.

3.2.1 Acousto-optics as a guide-star method

In that context, acousto-optics offers a local interaction between light and sound inside scattering media. The predominance of ballistic regime for ultrasonic waves in hydrogels and biological media allows us to control the shape of this interaction ([Lerosey and Fink, 2013](#); [Ma et al., 2015](#)). Furthermore, it does not imply introducing a probe or markers inside the medium which makes it non-invasive and easily implementable. The selective observation of the tagged optical field offered by digital holography gives access to the quantity of light passing through the ultrasonic focus.

3.2.2 Optical phase conjugation and time-reversal

We have seen that exploiting the degrees of freedom offered by spatial modulation techniques allows for the compensation of scattering effects in complex media. In the nineties, Mathias Fink initiated time-reversal techniques ([Fink, 1992, 1997](#)). First developed in acoustics ([Prada et al., 1996](#)) at the ‘waves and acoustics’ laboratory— co-ancestor, with the optics laboratory, of the Langevin institute at the ESPCI — the technique has been transposed to other domains of wave physics; microwaves ([Lerosey et al., 2007](#); [Fink et al., 2013](#)), optics ([Popoff et al., 2011](#)) and offered multiple applications especially for imaging and telecommunications. The technique is based on the reversibility of the wave equation, which means a propagation of a wave is unchanged by reversing the course of time. It is also inspired from the Helmholtz-Kirchhoff theorem. The Helmholtz-Kirchhoff theorem states that the simultaneous knowledge of a field and its derivative at all points of a closed surface, allows retrieving the field at any point within the delimited volume. Let us imagine a point source emitting a temporally-short wave, thanks to the Helmholtz-Kirchhoff theorem, if we install a continuous array of sensors around the source recording the passing of the wave, temporally-reversing the signal and playing it back, the sum of all the re-emitted wave will focus at the position of the source, both spatially and temporally. Experimentally, such an experiment is hard to implement. Indeed, to have access to a closed surface around sources with transducers spaced by half the wavelength, the total number of sensors would be tremendous. Moreover, the spatial distribution of the sensors provides discrete spatio-temporal sampling. In practice, we only have access to the field on a limited area, limiting the effective numerical aperture of the system. In monochromatic optics, the time reversal operation corresponds to conjugating (inverting) the phase of the optical field. It allow a spatial focusing of the light. In the next section, we see how photorefractive crystals can be used

to conjugate a complex optical field in order to focus light within a scattering medium.

3.2.3 Light focusing using phase conjugation

In this part, we present two techniques that allow performing optical phase conjugation. The first one, called wavefront adaptive holography, is based on the photorefractive effect in some crystals.

Photorefractive crystals: wavefront adaptive holography

Photorefractive crystals allow the phase conjugation by wavefront adaptation. These materials have the specificity to be sensitive to intensity through photorefractive effect. When subjected to a coherent light, the interference pattern creates dark and bright areas inside the material. This illumination pattern induces charge displacements inside the material. This spatial charge field, via the electro-optic effect, causes a change of the optical index proportional to the local optical intensity. This refractive index grating that is formed follows the light interference pattern with a spatial shift of $\lambda/4$. Depending on the crystal and the illumination power, the hologram inscription takes a fraction of a millisecond to 100 ms (0.3 ms to 100 ms in GaAs or BSO). This time is called the response time of the crystal. The hologram is then read by a reference plane wave. The holographic detection is thus made in two steps; by photorefractive effect, the interferogram of the signal beam and the pump beam imprints a weak modulation of the local refractive index inside the crystal. This phase grating thus created is read by a second pump beam in the opposite direction, see [Figure 3.6](#). The two pump beams have higher intensities than the incident signal beam. This interaction, called four-wave mixing, results in a diffracted beam whose field is proportional to the complex conjugate of the incident signal beam ([He, 2002](#)). This technique is called wavefront adaptive holographic because the wavefront of the reference beam is shaped by the index grating inscribed in the crystal. Photorefractive holography have been developed in acousto-optics imaging by François Ramaz at the the optics laboratory of the ESPCI and then at the Langevin Institute ([Ramaz et al., 2004](#); [Gross et al., 2005b](#); [Farahi et al., 2010](#); [Laudereau et al., 2015](#); [Bocoum et al., 2020](#)). Thus, four-wave mixing in photorefractive crystal allows complex conjugating an optical field. We see how this technique has been used for focusing into scattering media.

Time-reversal ultrasonically-encoded (TRUE) light imaging

Using phase conjugating, [Xu et al. \(2011\)](#) and [Liu et al. \(2015\)](#) proposed a method to focus inside a scattering sample using the acousto-optic effect. Their method is called time-reverse ultrasonically-encoded (TRUE) light focusing through scattering media. It consists in illuminating a scattering medium behind which an acoustic pulse is focused. After the limpid hydrogel, where the acousto-optic tagging occurs, the optical beam is multiply scattered a second time. The acousto-optical contribution of the final scattered field is focused back through the second scattering layer using wavefront adaptive

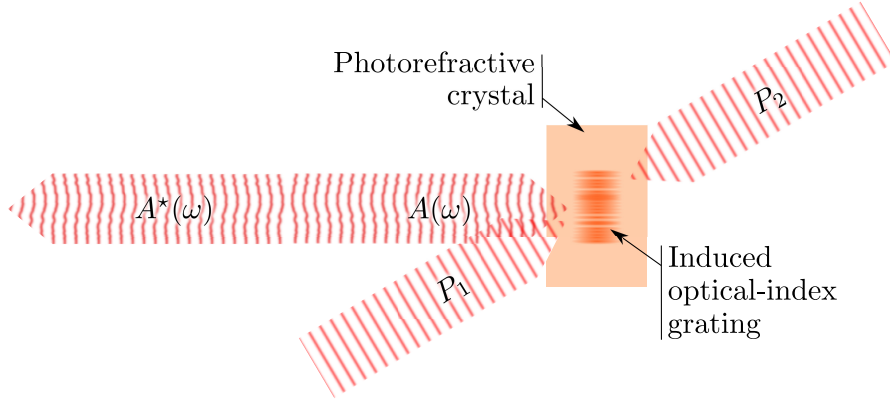


Figure 3.6 – Schematic of four-wave mixing in photorefractive crystals. The monochromatic beam $A(\omega)$ (signal) interferes with a first pump beam P_1 . The optical index grating originated from the fringe pattern then diffracts a second pump beam P_2 . The field of the order of diffraction created is proportional to the complex conjugate of the signal $A^*(\omega)$.

holography.

The schematic principle of the experiment is depicted on Figure 3.7. The first scattering layer consists in a ground glass diffuser, which produces static scattering. The second is intralipid-gelatin hydrogel, a scattering medium undergoing temporal decorrelation by translating it at a controlled speed. The thickness of the hydrogel of 1.5 mm approximately equals the transport mean free path $l^* \approx 1$ mm for a reduced scattering coefficient $\mu'_s = 1 \text{ mm}^{-1}$, corresponding to the simple scattering regime. The output optical field is directed towards a tellurium-doped tin thiohypodiphosphate $\text{Te:Sn}_2\text{P}_2\text{S}_6$ (SPS) photorefractive crystal. Two pump beams at the frequency of the positively-tagged photons are used to generate the time reversal of the acousto-optic signal. As the acousto-optic signal originates from the acoustic focal spot, the time-reversed field focuses back to this area. By conjugating the ultrasound plane with the sensor of a camera through a lens, the authors measured the optical focus. They obtain a size of half a millimeter which is consistent with the area of the acoustic focal spot at 3.6 MHz. The focusing has been possible even under a controlled decorrelation process. In order to simulated a decorrelation, the authors put the gelatine gel onto a motorized translation stage. Tuning motion speed allows directly to control the speckle decorrelation time. They manage to keep the accuracy of focusing with a short decorrelation time of 5.6 ms. Aiming to test *in vivo*, the experiment is repeated replacing the scattering hydrogel in translation by a living-mouse ear. This living tissue undergoes temporal decorrelation at a millisecond pace due to blood and lymphatic circulation and breathing movements of the mouse. Therefore optical phase conjugation does not manage to focus through the ear. This study is a milestone on the way of time-reversal optical focusing in living tissues.

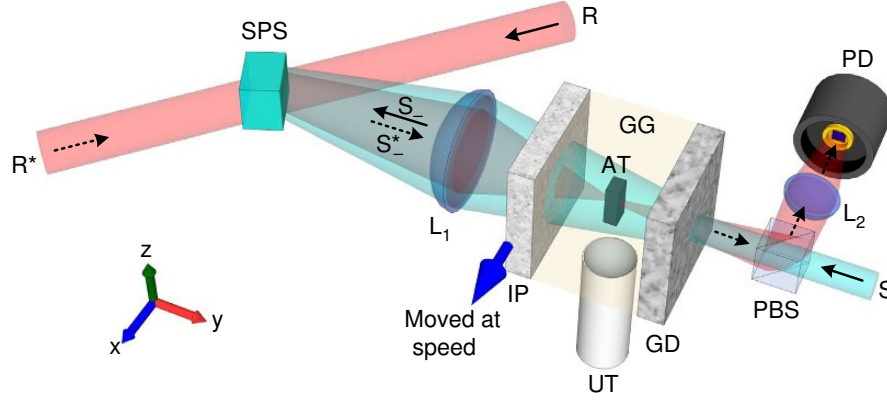


Figure 3.7 – Optical schematic for focusing inside a scattering medium by time-reversing the acousto-optic field from [Liu et al. \(2015\)](#). AT absorptive target, GD ground-glass diffuser, GG gelatine gel, IP Intralipid-gelatin phantom, L lens, PBS polarized beam-splitter, PD photodiode, R reference beam, R* reading beam, S sample light, SPS photorefractive crystal, UT ultrasonic transducer.

3.2.4 Digital optical phase conjugation (DOPC)

The phase conjugation has also been realized digitally using a SLM in combination with a camera. In 2012, [Wang et al. \(2012\)](#) focused an optical beam into a scattering medium using digital phase conjugation. The principle of the operation is very similar to four-wave mixing in photorefractive crystals. A signal beam illuminates the scattering medium in which the authors focused an acoustic pulse. The scattered field is then measured on a CMOS camera using a reference arm. The phase difference between acousto-optic contribution and the reference beam at $f_L + f_{US}$ is isolated using digital phase-shifting holography ([Yamaguchi, 2006](#)), see the optical schematic of the experiment on [Figure 3.8](#). Then the phase conjugated mask is numerically generated and displayed on a SLM. The SLM is illuminated with a plane wave, playing the role of the second pump beam in the photorefractive experiment. As in photorefractive process, the method benefits from the amplification of the tagged field provided by the reference pump beam.

Following this proof of principle of light focusing by digital optical phase conjugation (DOPC), [Ruan et al. \(2015\)](#) have proposed an iterative implementation of the TRUE method, called iTRUE. The principle of the method consists in implementing DOPC on the TRUE imaging setup of [Xu et al. \(2011\)](#), thus replacing the photorefractive holographic optical phase conjugation. Therefore, they focus a DOPC acousto-optic field through a scattering medium. As in ([Liu et al., 2015](#)), the width of focused beam corresponds to those of the acoustic focal area used to tag the primary optical beam. Then, the iterative process allows them to sharpen the optical focusing.

The focused ultrasonic excitation is repeated in order to acousto-optically tag the

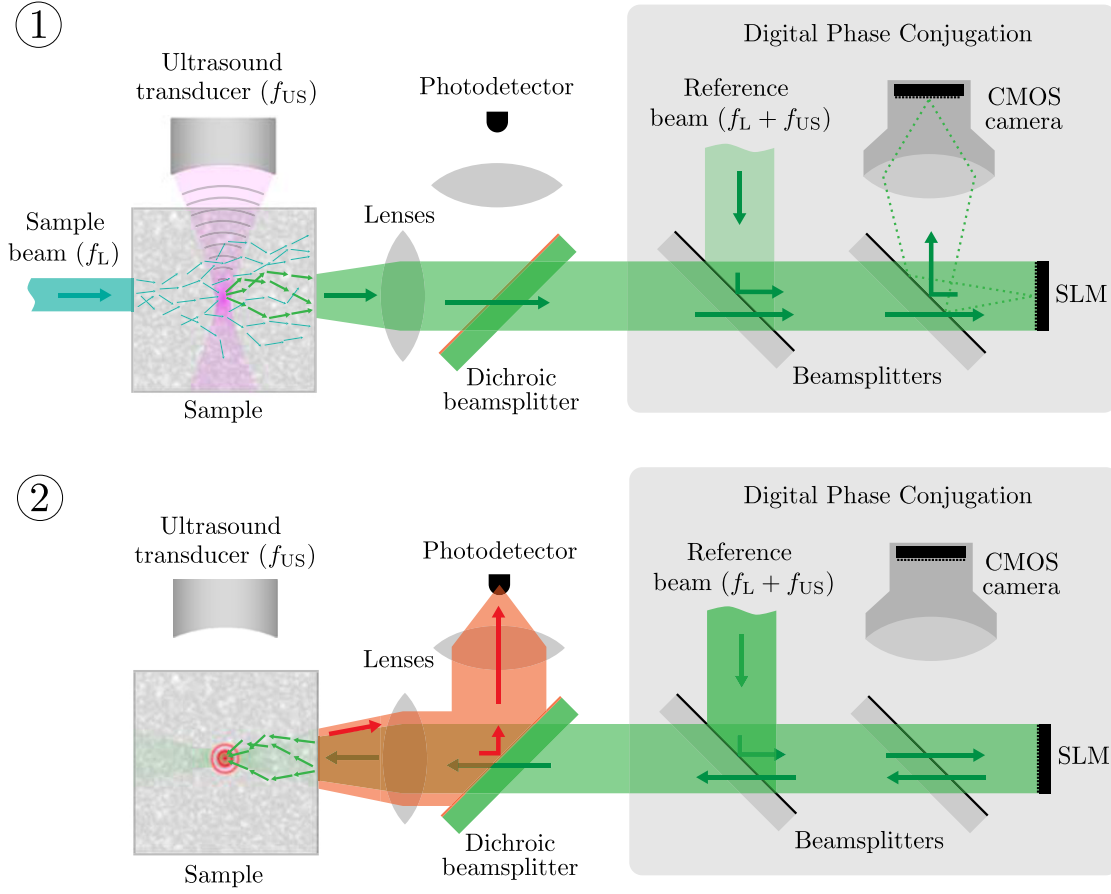


Figure 3.8 – Digital optical conjugation schematic adapted from (Ma et al., 2015). The signal beam illuminates the turbid sample where an ultrasonic pulse is focused. The scattered beam is directed toward the CMOS camera via a reflection on the SLM display. Here it interferes with the reference beam at $f_L + f_{US}$. This allows isolating the acousto-optic contribution and retrieving its phase. The opposite of the obtained phase mask is then displayed on the SLM. The reference beam is modulated by the SLM before entering in the scattering sample. Because of the digital phase conjugation, it focuses at the position of the acoustic focal spot. In order to monitor the efficiency of focusing, fluorescent beads are placed at the acoustic focal spot. The intensity of the fluorescence signal is captured by a photodetector.

focused beam, obtained with the first DOPC-TRUE step. Then, the tagged-photon contribution of the backscattered field is used to reproduce an DOPC-TRUE step. As the magnitude of the tagging spatially follows the amplitude of the acoustic focal area, the contribution of the photons tagged at the center weighs more than those tagged around.

As a consequence, the next DOPC-TRUE step produces a sharper light focusing. After N iterations, [Ruan et al. \(2015\)](#) reduce the optical TRUE-focal area by a factor $\sqrt{N}$. This method proves that one can overcome the AOI resolution imposed by the acoustic-diffraction limit. In the next chapter, we see how matrix approach of acousto-optic imaging also allows overcoming this limit.

3.2.5 Matrix approach of time-reversal

Focusing through a scattering medium using an iterative algorithm is efficient but requires a new calibration for each target point. For a linear medium, the relation between the optical field in a given plane and the one in an output plane can be described by a matrix. Such a matrix, called the transmission matrix (TM), was measured for the first time in [\(Popoff et al., 2010a\)](#). The knowledge of this matrix allows focusing on any target at the output and reconstructing an unknown image through a scattering medium [\(Popoff et al., 2010b\)](#).

The TM is the mathematical object used to describe mesoscopically the linear relation existing between the system input and output optical fields. It is defined at a specified wavelength. This matrix $\mathbf{T}$, composed of complex elements $t_{m,n}$, connects in amplitude and phase the n^{th} output component to the m^{th} input component (see [Figure 3.9](#)). For

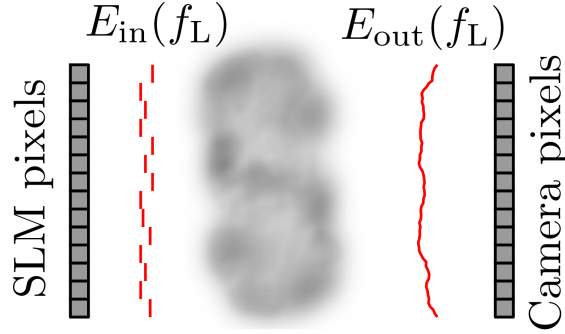


Figure 3.9 – Schematic of the transmission matrix principle. The scattering medium is illuminated by a monochromatic (frequency f_L) wavefront spatially-modulated by a SLM display. The output scattered field is sampled by the pixels of the camera.

a given input field E^{in} , the components of the output field E^{out} reads:

$$E_m^{\text{out}} = \sum_n t_{n,m} E_n^{\text{in}}, \quad (3.2)$$

with E_n^{in} and E_m^{out} respectively the complex amplitude of the optical field of the n^{th} input mode and the m^{th} output mode. In a system where light undergoes multiple scattering, the transmission is well represented by a complex random matrix [\(Goetschy and Stone, 2013\)](#). In an experimental context, the input and output basis for representing the fields needs to be wisely chosen to be conveniently implemented on the SLM and efficiently

detected. The knowledge of the TM for a temporally stable system allows controlling the output optical field by injecting the corresponding input wavefront.

The time-reversal operator

We have seen that phase conjugation allows back-propagating the optical field to its source it originates from. If we express it using the matrix formalism, the output field E_{out} produced by the point source E_{in} reads:

$$E_{\text{out}} = {}^t\mathbf{T} E_{\text{in}}, \quad (3.3)$$

where ${}^t\mathbf{T}$ denotes the transposed matrix of $\mathbf{T}$. Then, the time-reversal field E_{TR} produced by complex conjugating the field reads:

$$E_{\text{TR}} = \mathbf{T} E_{\text{out}}^*, \quad (3.4)$$

$$= \mathbf{T} \left({}^t\mathbf{T} E_{\text{in}} \right)^*, \quad (3.5)$$

$$= \mathbf{T} {}^t\mathbf{T}^* E_{\text{in}}^*. \quad (3.6)$$

We identify the hermitian conjugation denoted by the symbol $\dagger$:

$$E_{\text{TR}} = \mathbf{T} \mathbf{T}^\dagger E_{\text{in}}^*. \quad (3.7)$$

Therefore, the complex conjugated wavefront focuses onto the initial point source if $E_{\text{TR}} \approx E_{\text{in}}^*$. This condition is fulfilled if $\mathbf{T} \mathbf{T}^\dagger$ is close to the identity matrix. The operator $\mathbf{T} \mathbf{T}^\dagger$ is called the time-reversal operator (TRO). This is a square matrix operator expressed in the same basis as the input of the transmission matrix. This operator is experimentally used to invert the propagation for a measured transmission matrix. Indeed, contrarily to regular matrix inversion by $\mathbf{T}^{-1}$, this process avoids divergence issues due to noise measurement in experimental transmission matrix (Popoff et al., 2010b).

3.2.6 Measuring a transmission matrix

In 2010, Popoff et al. (2010a) experimentally measured the TM of a multiple scattering white paint layer using spatial light modulation. Indeed, as we have seen in the previous section, wavefront shaping techniques allow controlling a high number of degrees of freedom. Experimentally, the TM links the n^{th} pixels of the camera to the m^{th} pixel of the SLM. Because of the linearity of the system, the TM can be measured in any basis. As the SLM allows only the control of the phase, we cannot use the canonical basis of the SLM pixel. We chose the Hadamard vectors as an input basis for its noise robustness and its high contrast modulation.

The Hadamard matrices is defined as an orthogonal and symmetric matrix of size $2^n \times 2^n$ ($\mathcal{S}_{2^n}(\mathbb{K})$) built recursively:

$$\mathbf{H}_n \in \mathcal{S}_{2^n}(\mathbb{K}),$$

$$\mathbf{H}_0 = 1 \quad \mathbf{H}_1 = \begin{pmatrix} \mathbf{H}_0 & \mathbf{H}_0 \\ \mathbf{H}_0 & -\mathbf{H}_0 \end{pmatrix} \quad \cdots \quad \mathbf{H}_n = \begin{pmatrix} \mathbf{H}_{n-1} & \mathbf{H}_{n-1} \\ \mathbf{H}_{n-1} & -\mathbf{H}_{n-1} \end{pmatrix}.$$

Moreover, the Hadamard matrix is orthogonal, thus its inverse is proportional to its transpose conjugate:

$$\mathbf{H}_n^t \mathbf{H}_n = 2^n \mathbf{I}_{2^n}.$$

The complex elements of these matrices can be seen as binary phase values 0 and π , which can be easily implemented on the macro-pixels of the SLM (see Figure 3.10). Moreover, neglecting the diffraction effects, the global input power sent to the sample is conserved through the different vectors of the basis. An other interesting property is that the whole surface of the layout is used for each vectors, maximizing the input power and thus the measurement signal to noise ratio. An example of the Hadamard basis and its implementation on the SLM macro-pixels layout is presented in Figure 4.2.

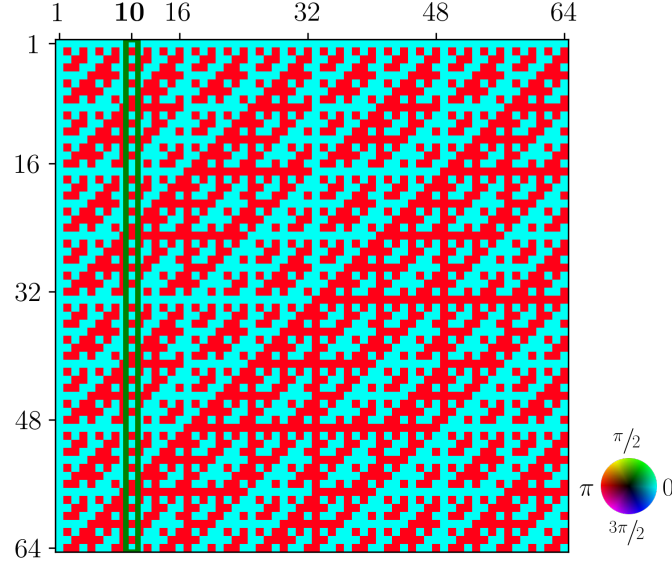


Figure 3.10 – Hadamard basis used on the SLM to measure the acousto-optic transmission matrix. The 10th column of the matrix circled in green corresponds to the vector used in Figure 4.2.

3.3 The acousto-optic transmission matrix

In a typical acousto-optics experiment, as presented in the chapter 2, a monochromatic input optical field at the frequency f_L illuminates an inhomogeneous medium. A focused ultrasound beam shifts the frequency of a portion of the photons ($f_L + f_{US}$). In transmission, both modulated (tagged) and unmodulated photons are present. The acousto-optic transmission matrix (AOTM) represents the linear link between the input

field at frequency f_L and the tagged photons at frequency $f_L + f_{US}$ (see Figure 3.11).

$$E_m^{\text{out}}(f_L + f_{US}) = \sum_n t_{m,n} E_n^{\text{in}}(f_L). \quad (3.8)$$

The tagged photons necessarily originate from the ultrasound region. Thus, compared to

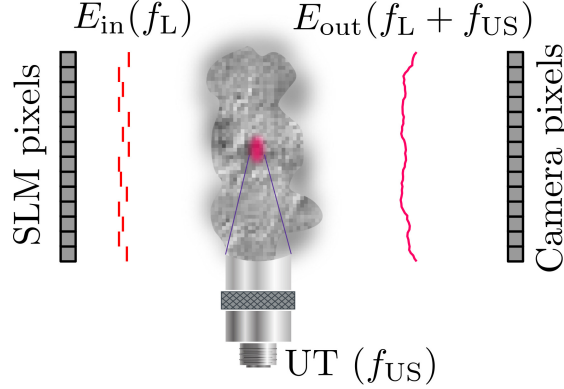


Figure 3.11 – Schematic the acousto-optic transmission matrix principle. The scattering sample is illuminated by a monochromatic (frequency f_L) wavefront spatially-modulated in phase by the SLM display. Throughout its propagation in the medium, a part of the photons are acousto-optically tagged by an ultrasonic wave at frequency f_{US} . The digital-holographic detection selects the ultrasound-modulated signal (at frequency $f_L + f_{US}$) which is sampled by the camera pixels. The acousto-optic transmission matrix of the sample is measured by recording all the output fields corresponding to the elements of the measuring basis, displayed by the SLM.

the full TM, the AOTM contains information about a subset of all the possible channels, the ones crossing the acoustically modulated region. The acoustic modulation is not uniform in the insonified area, all the channels do not bring an equivalent contribution to the transmitted speckle. As we have seen in the [chapter 2](#), the acoustic pressure profile has a maximum at its center and is well approximated by a gaussian shape. The shape of the selection along the acoustic-propagation axis depends on the modulation applied to the ultrasounds. For random phase jumps, we get a triangular shape. For chirped signals, we have a sinc function. In both cases, as the tagging efficiency is proportional to the amplitude of the pressure wave, a larger portion of the photons passing through the center of the acoustic spot is tagged. As a result, the channel passing through the center of the acoustic spot has a greater contribution to the speckle recorded by the camera. [Katz et al. \(2019\)](#) proposed to analyse the AOTM to reveal the transmissive channel passing through the center of the acoustic spot. Indeed, guiding light through these channels offers a better focusing technique than standard time-reversal technique in terms of resolution. The tool chosen for analyzing the AOTM is the principal component analysis. We present it in the next section.

3.4 The singular value decomposition

The singular value decomposition (SVD) is a powerful tool in matrix analysis. It consists in a decomposition that generalizes the diagonalization for cases when the input and output spaces are not the same. In particular, it works for non rectangular matrices. This is well suited for TMs as the input and output plane are at different positions. Let us consider $\mathbf{M}$ a rectangular transmission matrix of shape $M \times N$. The SVD factorizes $\mathbf{M}$ into:

$$\mathbf{M} = \mathbf{U} \mathbf{S} \mathbf{V}^\dagger. \quad (3.9)$$

The term $\mathbf{S}$ is a rectangular diagonal matrix of size N with real positive elements on the diagonal called the singular values λ_i . The singular values are sorted in decreasing order. Its index is defined as an eigen-channel index.

In the case of optical transmission matrices, the eigen-values of the TRO ($\mathbf{T} \mathbf{T}^\dagger$) correspond to the square of the singular values. The eigen-values of $\mathbf{T} \mathbf{T}^\dagger$ is the expected transmittance of the corresponding eigen-channels. Also, $\mathbf{U}$ and $\mathbf{V}$ are unitary matrices. Their columns correspond to the transmission eigen-channels, respectively at the output and the input planes, expressed in the canonical bases of the camera and SLM pixels.

SVD is a recurrent tool used in speckle engineering. It has been used for selective focusing (Popoff et al., 2011; Chaigne et al., 2014; Jeong et al., 2018), to identify open and closed channels (Vellekoop and Mosk, 2008; Kim et al., 2012; Yu et al., 2013) and to find dispersion-free states from the spectral version of the TM (Xiong et al., 2016; Ambichl et al., 2017). In the next chapter, we will see how the SVD of the acousto-optic TRO allows focusing through a scattering layer.

3.5 Conclusion

In this chapter we have reviewed wavefront manipulations implemented within the last two decades. We have seen that time-reversing the optical field allows focusing back to the emitters. Optical phase conjugation in photorefractive crystals is a monochromatic implementation of time reversal. Combined with acousto-optic tagging they have allowed focusing within a scattering sample in an area of size of the order the ultrasound wavelength. Then, the development of digital spatial-light modulators offers to control the input illumination in phase or in amplitude. These devices allow digitally building a matrix approach of optical system by characterizing their input and output modes. Once built, transmission matrices give access to the time-reverse operator.

We have also presented the singular value decomposition which, applied to an optical system, exhibits the input wavefront that maximize the optical transmission through the sample. In the next chapter, we present the experimental optical setup that allows measuring the acousto-optic transmission matrix. Then we see how the singular value decomposition can be harnessed to focus inside the acoustic focal area.

CHAPTER 4

Measuring the acousto-optic transmission matrix

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As mentioned in the previous chapter, in a scattering medium with an ultrasound excitation, the AOTM links the input optical field to the output acousto-optically tagged field. In this chapter, we present the experimental optical setup that allows measuring the AOTM. After introducing the AOTM measurement using random-phase-jump acoustic modulation, we see how the SVD applied to the AOTM allows focusing with a size smaller than the dimension of acoustic focal spot. Indeed, the resolution acousto-optic imaging is limited by the acoustic diffraction, i.e. a fraction of a millimeter with a few megahertz excitation, whereas the limit of optical focusing is the optical wavelength ($< 1 \mu\text{m}$). Hence, focusing beyond the acoustic diffraction limit would allow resolving more details in the tissue and make acousto-optic imaging closer to the resolution standard in microscopy.

4.1 Measuring the AOTM

In this first section, we present the optical setup for measuring the AOTM.

4.1.1 The optical setup

The optical setup is based on an AOI setup on which an optical wavefront shaper modulates the probe incident to the scattering medium (see [Figure 4.1](#)). As for AOI, we use a continuous monochromatic laser source whose wavelength is in the therapeutic window ($\lambda = 780 \text{ nm}$). The acoustic modulation is provided by an single-element ultrasound transducer and a digital holographic detection setup measures the tagged-photon contribution of the output scattered field.

The scattering sample that we use is a ground glass diffuser and the ultrasonic wave is focused at the center of the scattered light beam in a water tank placed behind the diffuser. The probe beam is expanded and collimated in order to cover the whole surface of the SLM.

4.1.2 Wavefront shaping of the incident beam

As proposed by [Katz et al. \(2019\)](#), we measure the AOTM through Hadamard-vector basis. The surface of the SLM offers few millions of degrees of freedom with as many pixels. When we modulate the wavefront, pixels are actually grouped into macro-pixels with a chosen geometry. The overall lattice of macro-pixels can be chosen to design the geometry of the modulation and also to fit the desired number of degrees of freedom. To implement the hexagonal pattern to the SLM pixel array, we use a Python library developed by Sébastien Popoff ([Matthès et al., 2019](#)). The SLM display is controlled from a computer as a secondary screen. The target phase array, encoded on 8 bits pixel by pixel, is sent by the computer to the SLM controller. As we have seen in the previous section, reflection LCOS-SLMs are made of liquid crystal cells sandwiched between a reflective surface and a transparent electrode. This structure has two negative effects on the impinging light: first the pixel array acts as a two-dimensional diffraction grating, the diffraction angle is $\approx 3.5^\circ$ at 780 nm . Secondly, the glass surface of the

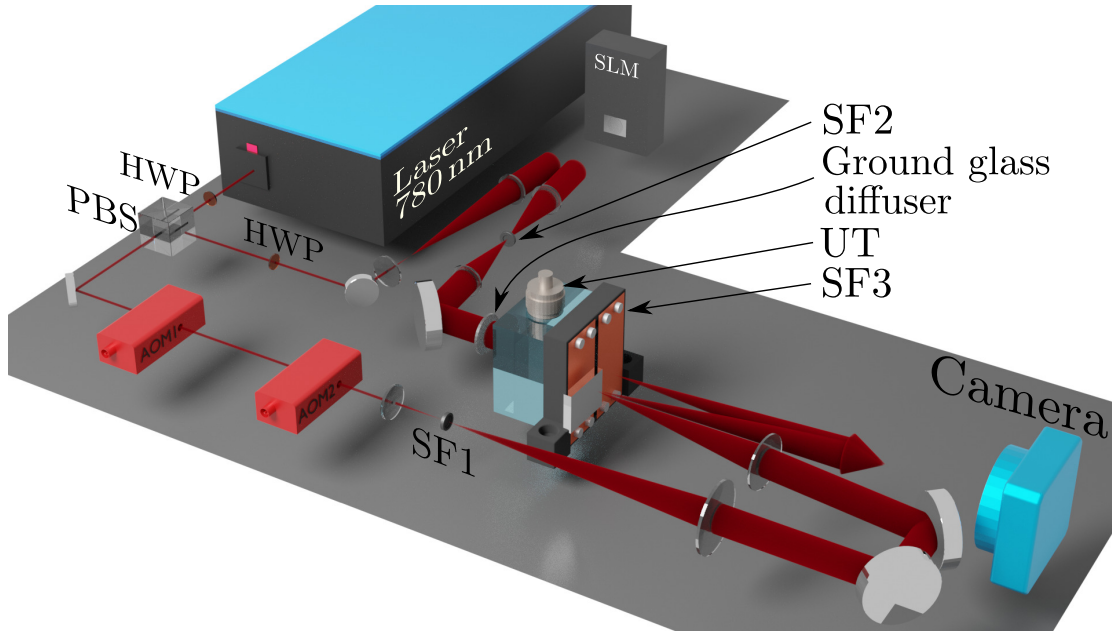


Figure 4.1 – Schematic of the optical setup used to measure the AOTM. The continuous laser at 780 nm is split in two beams through a polarized beam splitter, the power ratio between the two arms is adjusted with a half-wave plate (HWP). (SF#) Spatial filters: (SF1) is a pin hole with a 200 μm diameter, (SF2) is a width-adjustable slit and (SF3) is the slot used to limit the numerical aperture of the speckle for off-axis holography, (UT) ultrasonic transducer.

SLM adds a unmodulated reflection in the specular reflection direction. To get rid of the unmodulated light, a periodic phase ramp along one direction is superimposed to the desired pattern (see [Figure 4.2](#)). Thus, at the output of the SLM, a width-adjustable slit placed at the Fourier plane of a spherical lens allows selecting only the modulated first-order of diffraction.

4.2 The acoustic modulation

As we have seen in the [section 2.1.4](#), the use of random-phase-jump modulation allows improving the resolution along the acoustic propagation axis. It also improves the signal-to-noise ratio using a longer integration time. To match the same resolution as [Katz et al. \(2019\)](#), we first apply random-phase jumps on a 15 MHz carrier signal. After multiple tries, we did not manage to get sufficient signal to noise ratio to correctly retrieve the phase of the acousto-optic contribution. We hypothesized that the random phase jumps is too spectrally-large with respect to the linewidth of the ultrasonic transducer. Moreover, in order to avoid deterioration due to overheating one has to respect the power

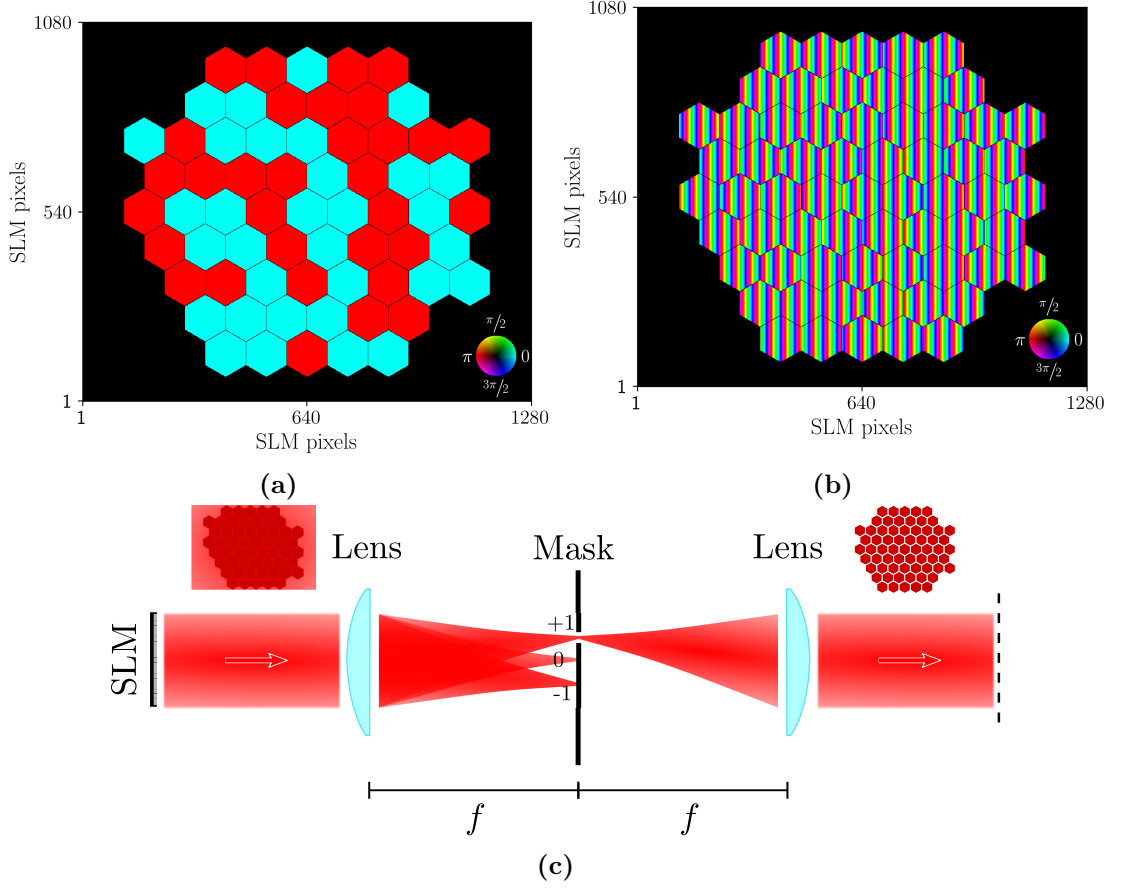


Figure 4.2 – Input spatial modulation with Hadamard vectors. Here the vector displayed corresponds to the 10th column of the Hadamard 64 matrix on [Figure 3.10](#). The 64 hexagons (a) corresponding to the 64 elements of the vector, pave the center of the SLM. This layout is generated by a Python library. A phase slope (reduced here for readability, the real slope is 8 pixels for 2π) horizontally modulates the layout. As a phase slope is superimposed specifically to the layout (b), the modulated contribution of the SLM output beam is spatially separated from the unmodulated specular reflection in the Fourier plane of a lens (c). A second lens with the same focal distance f allows imaging the SLM in the focal plane with a magnification of 1 (schematic inspired from ([Popoff, 2013](#))).

dissipation limit in the transducer (125 mW). As the temporal length of random phase jumps (2 ms) is a thousand times longer than pulses used by [Katz et al. \(2019\)](#), the input voltage of the signal had been reduced by a factor of ten. The quantity of tagged photons suffers from this reduced acoustic excitation and should be compensated by the longer integration time allowed by autocorrelation method as detailed in [section 2.1.4](#).

For these reasons, we decided to change for a 5 MHz probe that we have characterized

in chapter 2. As we have seen on Figure 2.2, its focal area is wider than the 15 MHz one:

$$W_{-6\text{ dB}} = 1.02 \frac{c_s F}{f D}, \quad (4.1)$$

with $W_{-6\text{ dB}}$ the -6 dB pulse-echo beam diameter, F the focal length, f the frequency and D the diameter of the membrane. Thus, using this probe while keeping the same jump width, we multiply the tagging area by nine with respect to the 15 MHz-probe focal area (see Figure 4.3). Using a width for random phase jumps of 200 ns, the acousto-

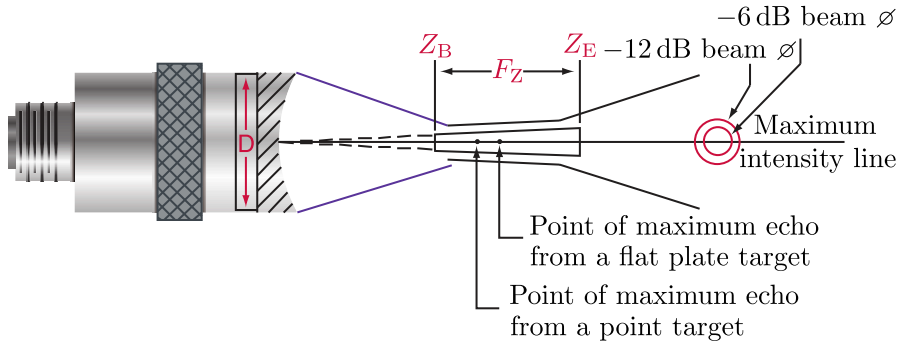


Figure 4.3 – Schematic of ultrasonic transducer focus and its focal length adapted from Olympus transducer technical file. The symbol $\varnothing$ denotes the diameter.

optic focal area is elliptic with a size of 0.3 mm along the acoustic column and a 0.75 mm FWHM in the transverse direction.

4.3 The necessity of automating the experiment

A large part of the work was devoted to built codes to handle the diversity of electrical devices used in the experiment (see Figure C.5 in section C.2). Indeed, as it combines two in-phase modulated optical beams interacting with acoustic emitters, the experiment had to obey a precise timing. To do so, we developed a Python library to manage each individual device, from delay generators to high-speed cameras, arbitrary waveform generators and the spatial light modulator. This task took into account the different language specificities of each devices and the global experiment was completely automated to go through measuring the acousto-optic matrix during a night. A schematic of the optical setup and graphical explanations of the electrical connections and the timing of signal executions are presented in the appendix section C.2.

4.4 Managing the sources of noise

In order to control the stability of the optical system, we measure the spatial correlation of the speckle as a function of time. Indeed, temporal decorrelation and spatial drifts of the observed patterns will be manifested by a change in the intensity map on

the camera sensor. The correlation between two $M \times N$ intensity images $A = a_{i,j}$ and $B = b_{i,j}$ of same shape is given by:

$$C(A, B) = \frac{\sum_{i,j} a_{i,j} b_{i,j}}{\sqrt{\sum_{i,j} a_{i,j} \sum_{i,j} b_{i,j}}}. \quad (4.2)$$

This allows monitoring long time shift due to thermalization of the optical components, especially within the first minutes of powering on the laser. Moreover, the water used to refill the water tank to compensate for the evaporation also needs to be at equilibrium with the room temperature to avoid optical index turbulence. Then, to get rid of decorrelation sources and lengthen the decorrelation time, we proceeded in successive steps:

- We begin by measuring the correlation on each arm. On the object arm we replace the SLM by a plane mirror to avoid eventual phase distortion or refreshing effects.
- Then, we checked the stability of all opto-mechanical component. We chose sturdier mounts, with wide basis, for crucial elements, such as the SLM and we clamped every wires to the optical table.
- We built an enclosure to isolate the setup from the flow of the air conditioning unit and placed any source of thermic noise outside (AOM amplifiers, main body of the fast camera) to prevent from thermic air flows across the optical paths.

Finally, we achieved maintaining 0.99 correlation of the intensity of the optical (without acousto-optic interaction) for few hours as shown on [Figure 4.4](#). However, a global shift

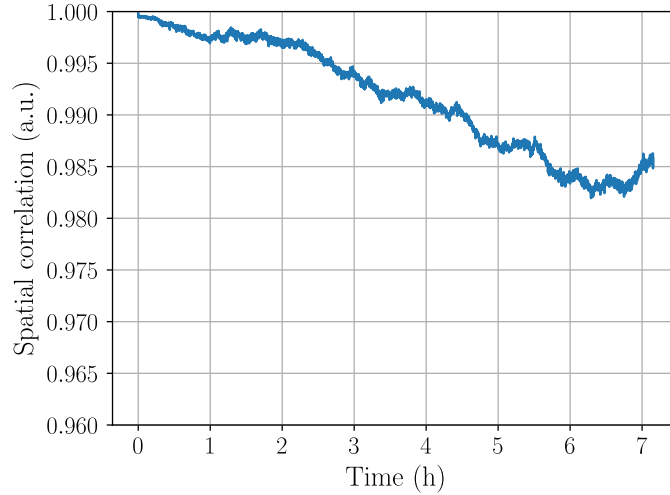


Figure 4.4 – Evolution of the spatial correlation of the interference pattern intensity through the complete optical setup.

can remain between two consecutive images. As the SLM is based on phase modulation, this could perturb wavefront shaping. In next section we will see how we get rid of phase uncertainties.

4.5 Building the AOTM

The transmission matrix is measured by displaying the different phase vectors corresponding to the N_{SLM} vectors of the input basis. In order to have an indicator of the stability and the reliability of the measurement, the acousto-optic field of the vector of reference, a plane phase modulation on the SLM, is measured before each Hadamard-vector all along the acquisition. Figure 4.5 shows the spatial correlation of the output speckles corresponding to all the reference vectors. These correlation data provides us two pieces of information. First, after a long acquisition sequence, it gives us an overview of the speckle stability. These reference flat phase vectors also allow compensating phase

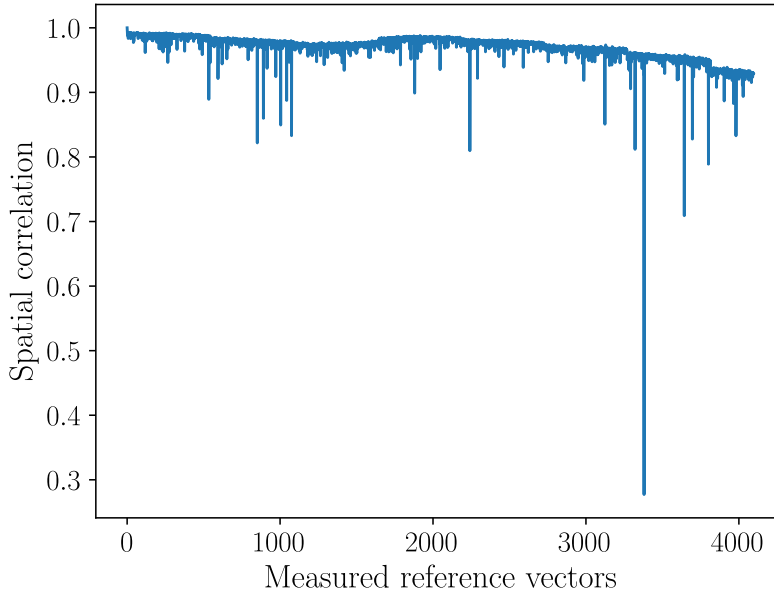


Figure 4.5 – Spatial correlation of a reference vector along the acquisition.

drifts. As they are displayed before each input mode injection, the phase of the output field measured $\phi_k^0(\eta, \xi)$ is then subtracted to the following Hadamard vector $\phi_k(\eta, \xi)$ output field.

$$E_k(\eta, \xi) = |E_k(\eta, \xi)| e^{i(\phi_k(\eta, \xi) - \phi_k^0(\eta, \xi))}. \quad (4.3)$$

Nevertheless, this process multiplies by two the total number of injected and measured fields and, as a consequence, the total number of frames recorded by the fast camera. As the limiting factor is the refresh rate of the SLM of 10 Hz, the total acquisition time is 20 min. Because of the size of the acquired images of 512×512 pixels, the internal memory of the camera of size 4 Gb needs to be completely unloaded periodically. This brings the total acquisition time to 122 min. The test of the internal memory capacity for each measurement and the slicing of the acquisition was included in the main driving

code to be automated.

The successive images, example presented on Figure 4.6, are then unfolded to be stack in an intermediate matrix $\mathbf{T}_H$. As it has been measured in the Hadamard vector basis we need to multiply it by the Hadamard matrix and normalize it:

$$\mathbf{T} = \frac{1}{2^N} \mathbf{H} \mathbf{T}_H. \quad (4.4)$$

The 4096-element basis matrix, i.e. implying 2^{12} degrees of freedom on the SLM, is presented on Figure 4.7.

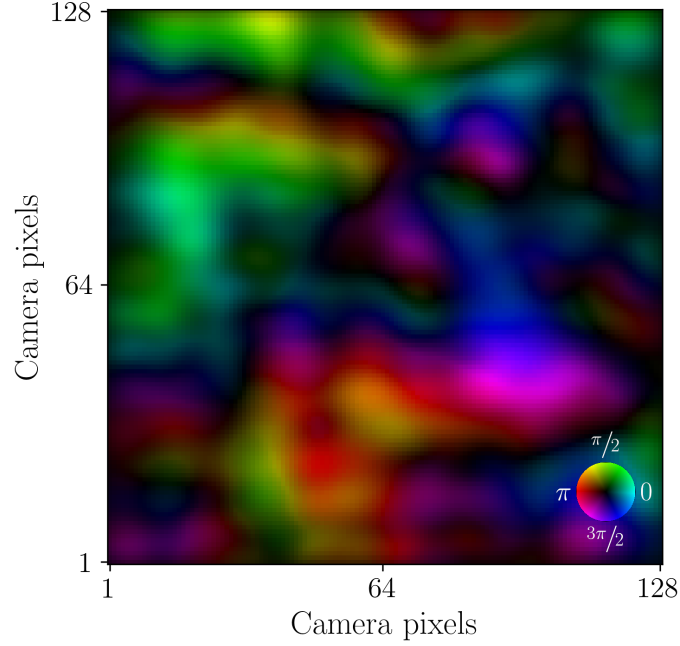


Figure 4.6 – Experimentally measured AO transmitted field.

Then, we compute the time-reverse operator:

$$\mathbf{O} = \mathbf{T} \mathbf{T}^\dagger, \quad (4.5)$$

of size 4096×4096 , linking the canonical SLM basis to itself through a round trip across the system (see Figure 4.8). As it has been built from the acousto-optic contributions, the elements $o_{i,j}$ of this symmetric square matrix carries the amplitude of the optical field coming from a segment i (or j) on the SLM, spatially-filtered at the ultrasound focus due to the tagging and the off-axis holographic selection — i.e. selecting only the component of the field tagged by the ultrasonic field — and then back-propagated onto the segment j (respectively i) of the SLM.

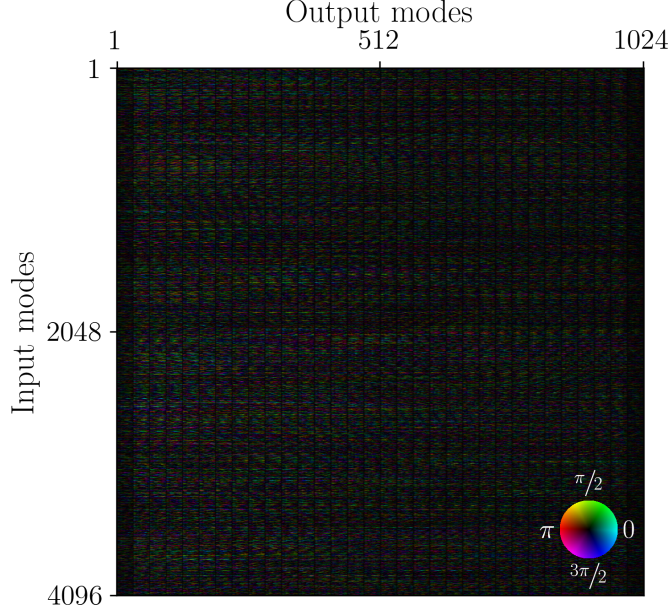


Figure 4.7 – An experimental AOTM measured in a 4096-element basis.

4.6 Light focusing inside the acoustic focal area using singular value decomposition

The singular value decomposition of a square matrix identifies the singular-vectors of the system and sort them in decreasing order of their corresponding singular values. Applied to the acousto-optic TRO, the singular vectors associated with the highest singular values represent the complex vectors, expressed in the canonical basis of the segments on the SLM, that conduct the highest number of photons to be tagged through the ultrasound focus. Conversely, the last phase-vectors, corresponding to the weakest singular value, provide the optical fields through which the acousto-optic interaction is the weakest. [Figure 4.9](#) shows the distribution of the normalized singular values $\tilde{\lambda}_i$ through their histogram and their value repartition. Singular values are normalized by the root of the sum of all the squared singular values, this sum corresponds to the total optical energy of acousto-optically tagged photons ([Katz et al., 2019](#)).

$$\tilde{\lambda}_i = \frac{\lambda_i}{\sqrt{\sum_j \lambda_j^2}}. \quad (4.6)$$

Once we have the singulars vectors, we can propagate them virtually through the system using the measured AOTM by computing the output field $E_{\text{out}} = \mathbf{T} E_{\text{in}}$. Thus, we calculate the acousto-optic field they would produced if displayed by the SLM. Furthermore, as the light propagation from the ultrasound plane to the camera plane is in free-space, we retrieve the field in the ultrasound plane directly by processing an inverse

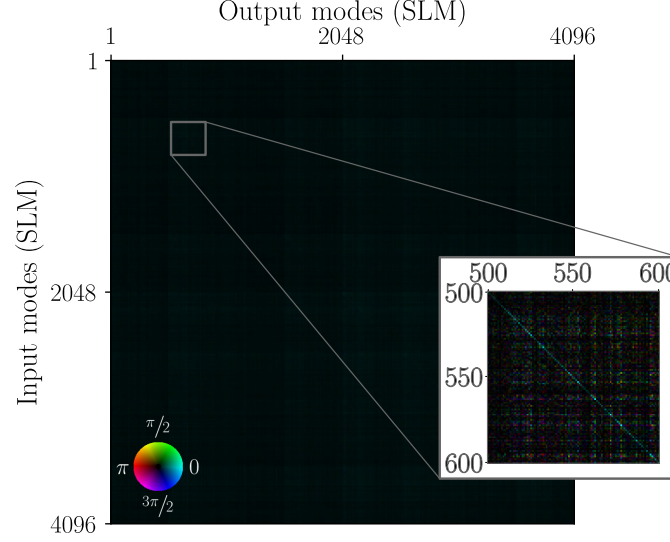


Figure 4.8 – The time-reversal operator corresponding to the experimentally measured AOTM shown on Figure 4.7.

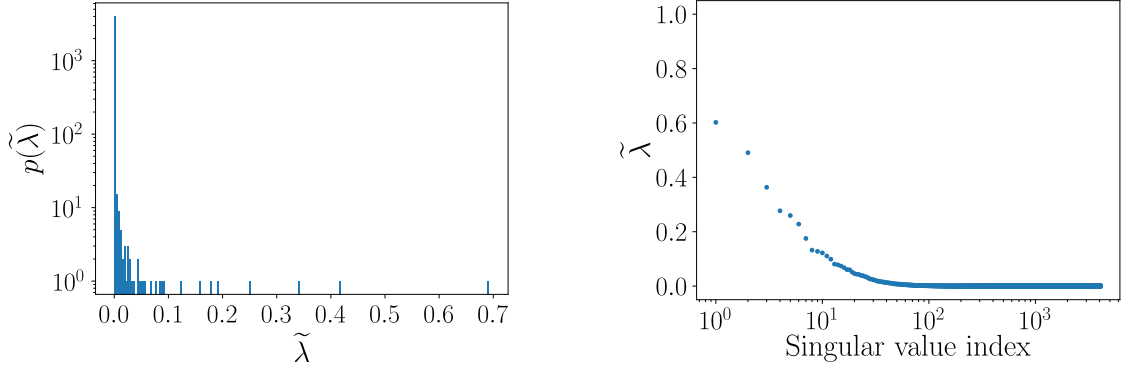


Figure 4.9 – Distribution of the singular values of the AOTM. (a) histogram of the distribution. The first singular values, are located on the right of the histogram whereas the last one, more numerous and with lower values are grouped on the left. (b) Value of the $\tilde{\lambda}_i$ sorted by decreasing order.

two dimensional Fourier transform. Figure 4.10 and Figure 4.11 present the computed complex field in the ultrasonic plane corresponding to singular vectors of increasing indices. As tagging efficiency increases as the square of the local acoustic field amplitude, the first singular vectors corresponds to wavefronts that focus light at the center of the acoustic spot. Indeed, at this point, the efficiency of the ultrasound tagging is maximal. The following singular vectors correspond to optical fields in the object plane that cover a larger area around the central spot of the first singular vector. Starting from index 100, the intensity of the illumination forms a ring-shaped pattern (see Figure 4.11). These

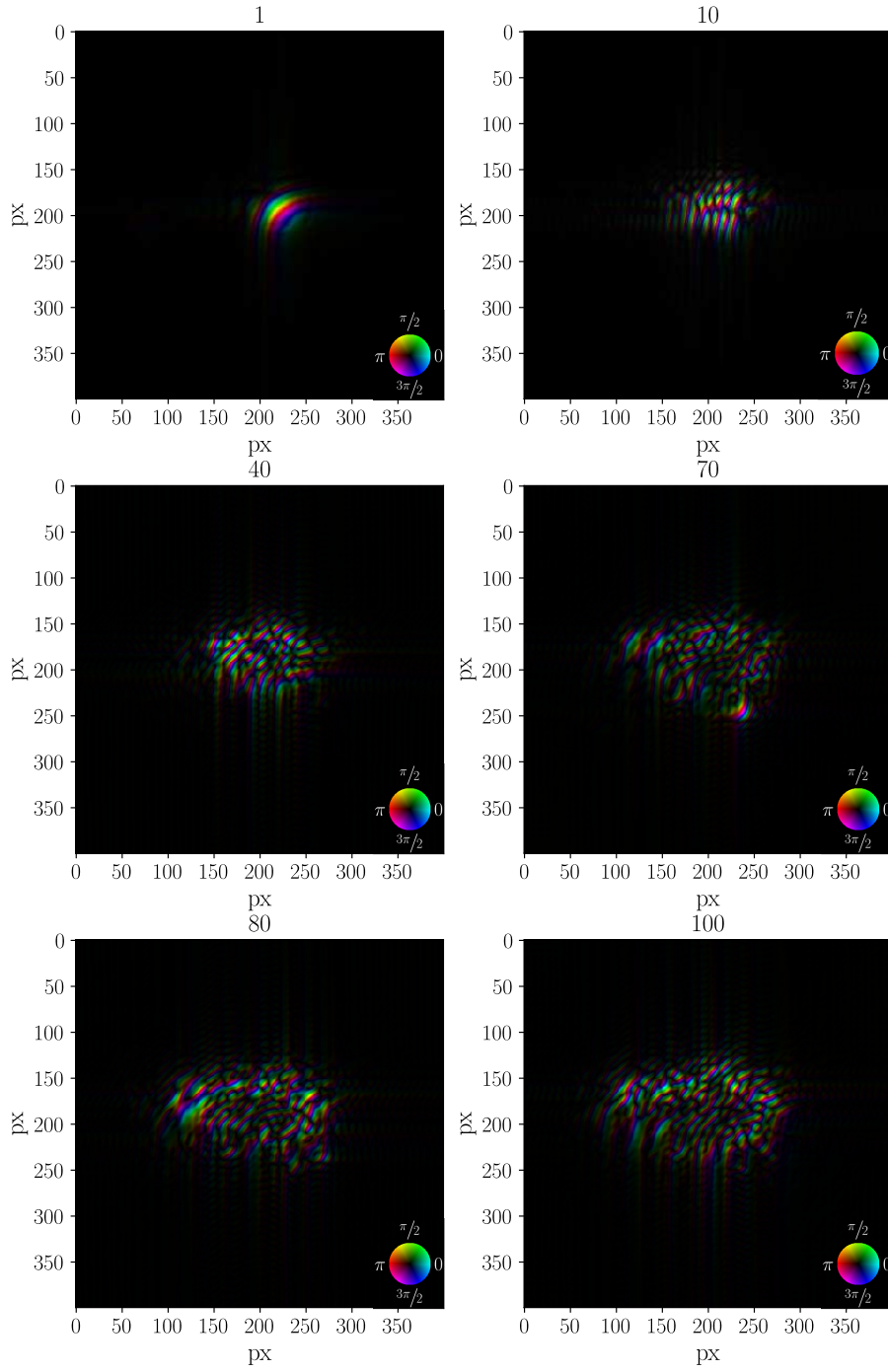


Figure 4.10 – First singular vectors. Optical-field profiles in the ultrasound plane corresponding to singular vectors associated with the singular values of index 1, 10, 40, 70, 80 and 100.

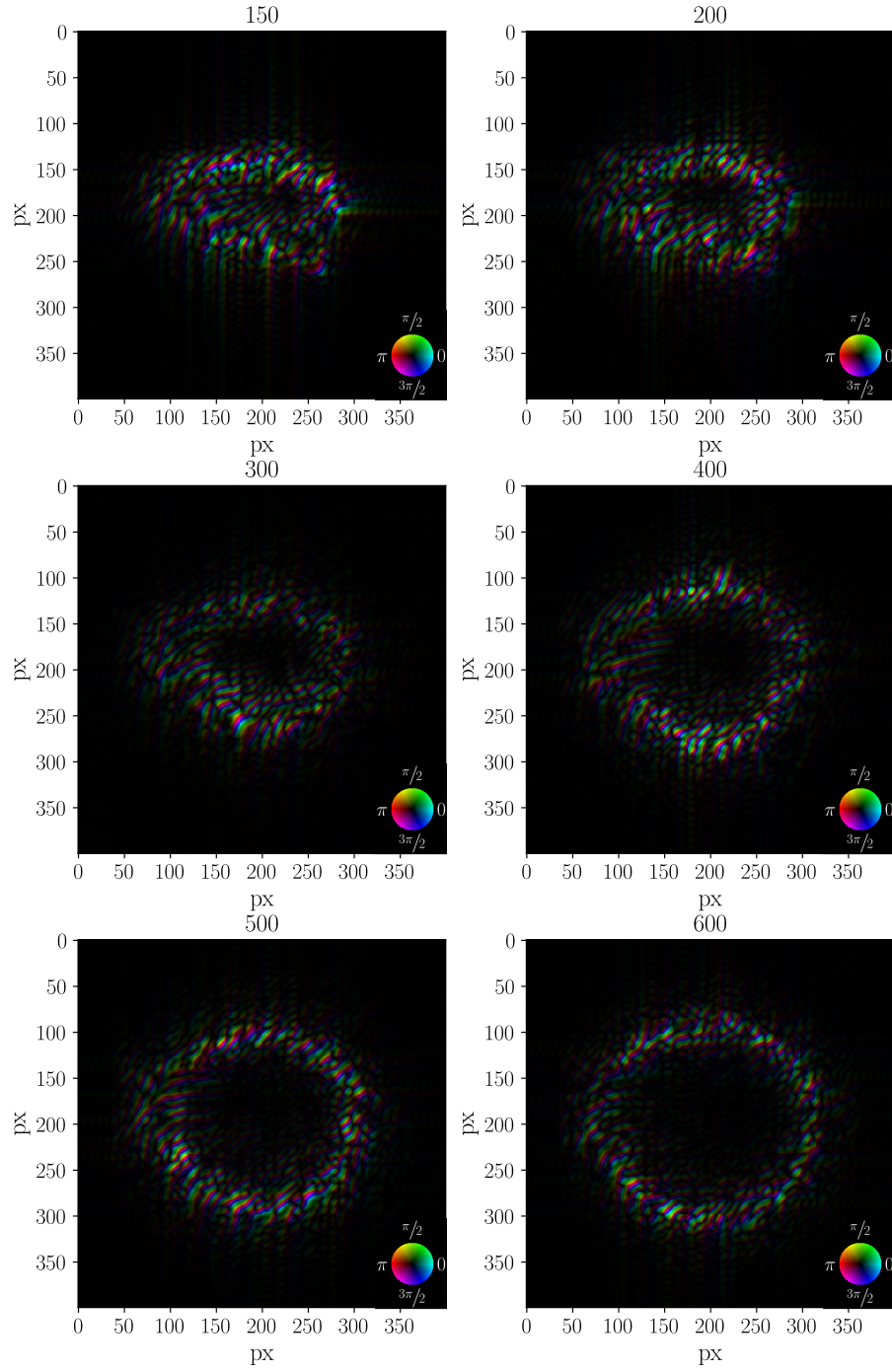


Figure 4.11 – High index singular vectors. Optical-field profiles in the ultrasound plane corresponding to singular vectors associated with the singular values of index 150, 200, 300, 400, 500 and 600

shapes also observed by [Katz et al. \(2019\)](#), correspond to the contour of the tagging function at a given amplitude. This effect starts at an index depending on the ratio between the number of speckle grains inside the ultrasound focal area and the number of degrees of freedom provided by the SLM modulation. [Katz et al. \(2019\)](#) observe the first rings starting around index 10. In our case, they appear around index 100 for a 4096-element basis. This difference could be explained by the different ultrasound focal areas between the 15 MHz used in [Katz et al. \(2019\)](#) and the 5 MHz that we use here. Indeed, the numbers of degrees of freedom necessary to address distinct modes inside the acoustic spot grows with the total number of speckle grain to address in the tagged volume. [Katz et al. \(2019\)](#) gives a demonstration of the number of degrees of freedom N_{SLM} necessary to address distinct regions of space with the two first singular vectors:

$$N_{\text{SLM}} > \frac{16\sqrt{2}}{\pi} \left(\frac{\sigma_{\text{US}}}{\sigma_{\text{speckle}}} \right)^2, \quad (4.7)$$

with σ_{speckle} the speckle grain size in the ultrasound plane and σ_{US} the size of the ultrasound focal spot. [Equation 4.7](#) shows that the degrees of freedom necessary to focus scales with the ratio of a speckle and the ultrasound spot areas. The maximum of the tagging function is quite flat due to the flat top of the envelope of the ultrasound focal shape. It implies that multiple speckle grains are located in this area and are tagged approximately with the same efficiency. Moreover, without this clear tagging amplitude criterion, the singular value decomposition does not achieve lifting the degeneracy between those approximately equivalent speckle grains in terms of tagging efficiency.

We also notice that our rings are narrower along the acoustic axis. This observation is compatible with the anisotropy of our selection area due to the use of random phase jumps. Indeed, we use phase jumps of duration $0.67 \mu\text{s}$, corresponding to an axial resolution of $100 \mu\text{m}$, while the transverse resolution is fixed by the waist of $\approx 100 \mu\text{m}$ at 15 MHz. Finally, the resolution of the focusing has to be compared with the efficiency of the TRUE method [Liu et al. \(2015\)](#). As this method consists in complex conjugating a random acousto-optic field, the focused zone is thus spread on the area of tagging, i.e. 0.75 mm . [Figure 4.12\(a\)](#) shows the intensity of the focused illumination produced by the injection of the first singular vector. The transverse FWHM of this focused illumination is $\approx 180 \mu\text{m}$. The resolution increase is similar to the one obtained by [Katz et al. \(2019\)](#). We then focused light on a spot 4 times smaller than the size of the ultrasound focal spot dimension. Moreover, the achieved resolution overcomes those obtained by [Ruan et al. \(2015\)](#) with iTRUE light focusing. The singular value decomposition directly provides the optimal illumination for focusing at the center of the acoustic focal area, which is achievable for iTRUE after an infinite number of iterations. This demonstrates that acousto-optics focusing and imaging is not fundamentally limited by the wavelength of the ultrasound excitation.

The [Figure 4.12](#) compares the experimental focusing, measured by a observation camera conjugated with the ultrasound plane, with the acousto-optic field, retrieved by computing the inverse Fourier transform on the field measured by the holographic detection. We observe that the first singular vector numerical projection (see [Figure 4.12\(a\)](#))

corresponds to actual field measured by the observation camera. The width of the optical focal spot is the same for the two images proving the focusing efficiency. We produce the same projection for the fiftieth singular vector on Figure 4.13 with the same resolution. However, the ring shape of higher order vectors do not appear. This could be due to errors in the AOTM measurement due to decorrelation of the medium. Because of the long measurement time, singular vectors injected after the measurement would lead to different outputs that the computed ones as the medium has changed over time.

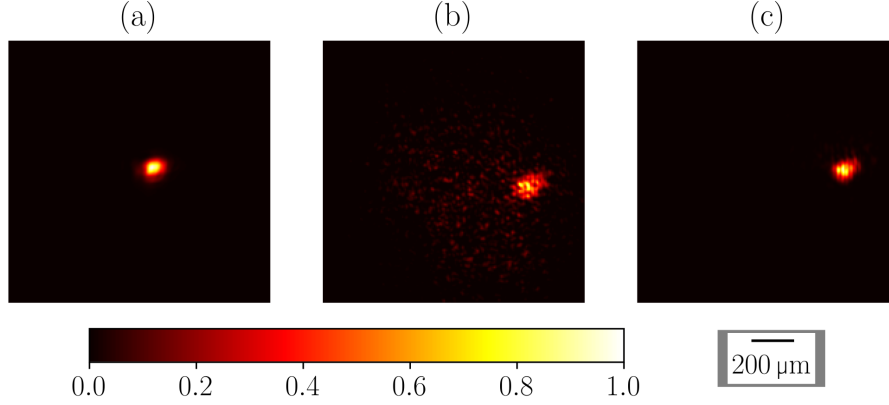


Figure 4.12 – Normalized intensity of first singular vector projections in the ultrasonic plane: (a) numerically using the measured AOTM, (b) physically through the system but only keeping the acousto-optic contribution, (c) physically through the system considering tagged and untagged light.

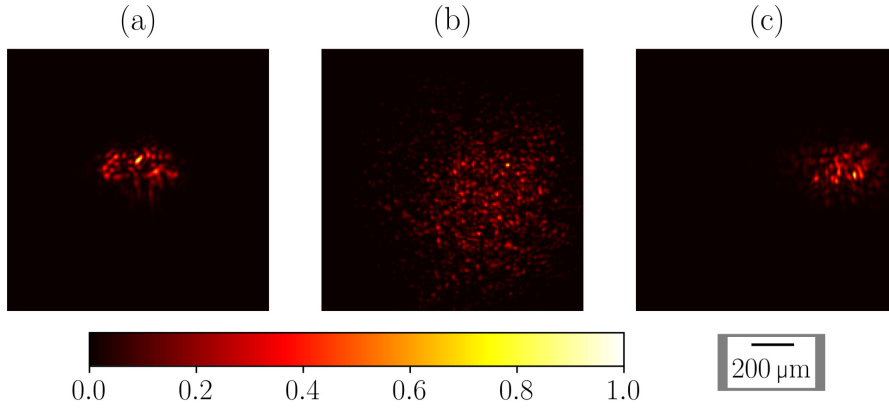


Figure 4.13 – Normalized intensity of the fiftieth singular vector projections in the ultrasonic plane: (a) numerically using the measured AOTM, (b) physically through the system but only keeping the acousto-optic contribution, (c) physically through the system considering tagged and untagged light.

4.7 Conclusion and perspectives

To conclude this experimental part of the AOTM measurement, we have seen how we measure the AOTM. Based on the off-axis detection of the acousto-optically tagged photons, the experimental setup requires reliable synchronicity and the phase matching over hours. Then, using the singular value decomposition applied on the time-reversal operator, method previously proved by [Katz et al. \(2019\)](#), we focus through a scattering layer beyond the acoustic diffraction limit imposed by the ultrasonic excitation. Thus, we achieve a 4 times narrower resolution than existing guide-star method based on acousto-optic coupling such as TRUE phase conjugation. Also, we reach the asymptotic focusing limit of the iterative TRUE method (iTRUE). Indeed, the first singular vector addresses the few optical speckle grains located at the center of the acoustic focal area. The higher-index singular vectors then address equivalent peripheral speckle grains creating ring shapes, following the geometry of the acoustic focal area. Selectively addressing the speckle grains following the excitation profile of the ultrasound offers degrees of freedom in the local illumination. Thus we can imagine a diversity of illumination profiles allowing multiple spatial sensing of the optical contrast. Furthermore, here we keep the single-element transducer excitation which geometrically constraints the shape of the acousto-optic tagging function. In future work, we could imagine using multi-element acoustic probe such as the echographic one used in [chapter 2](#) to provide more complex acousto-optic interaction.

IN this thesis, we have developed matrix approaches in acousto-optic imaging. These approaches rely on the wavefront shaping of the acoustical and optical waves. Here we summarize the principal results of this work.

After reminding how optical imaging techniques are crucial for the diagnosis and early detection, the chapter one has highlighted the limits of standard optical imaging techniques in biological tissues due to multiple scattering. Nevertheless, we have shown that diffuse optical tomography retrieves the absorption and scattering map inside the media solving the propagation inverse problem. But this all-optical method demands for high-computing resources and finding a compromise between depth of imaging and signal-to-noise ratio. Therefore, multi-wave techniques take advantages of the ballistic propagation of acoustic waves to probe the optical contrast in depth. The acousto-optic effect allows tagging light passing at a specified region and thus recovers an acoustic-diffraction resolution at centimeter depths. The amplitude of the tagged light is directly linked to the local contrast due to absorption and scattering. Spectral hole-burning meets the challenge of tagged-light detection both in reflection and detection and is getting closer to *in vivo* applications. The detection method that we use, off-axis digital holography allows retrieving the complex optical field which is a clue element to monitor the phase of the scattered wave.

The second chapter present different modulation strategies that have been developed for acousto-optic imaging. Temporal modulation of spatially focused ultrasound allows sectioning a depth narrower than the focusing area of the probe. Then transducer arrays allow spatially broad excitations, improving the signal to noise ratio. This improvement yields to a faster acquisition time compatible with imaging sessions in dynamic sample such as *in vivo* tissues. Furthermore, structuring the acoustic excitation spatially and temporally provides Fourier-transform imaging. This method consists in addressing the spatial Fourier components of the illumination plane using two-dimensional periodic pattern played by the ultrasound-scanner probe. Simulations on the acoustic distortions highlight frequency doubling in the Fourier domain which nevertheless limits the

possible structured excitations. These diffraction effects could be taken as advantages of multi-spectral excitation by measuring the tagging functions for each pattern in a known environment e.g. ballistic observation in a water tank. Then we could adapt the reconstruction taking into account all two-dimensional Fourier spectrum of the tagging pattern including effects of pattern edges and acoustic diffraction.

The next chapters have focused on the wavefront control of the input illumination. In the third chapter, we have introduced wavefront shaping techniques and their efficiency to compensate for the phase distortion applied by scattering. Then we have seen that the non-invasivity of acousto-optic tagging allows guide-star optimization. The measurement of the acousto-optic transmission matrix provides a global information on optical modes transmitted through the ultrasound focus. The singular value decomposition that we performed allows exhibiting the most effective input modes, identified as the eigen-vectors of the time-reversal operator. The spatial light modulation allows phase-shaping the input wavefront to address these modes. Therefore, the first singular vector of the time-reversal operator guides light at the center of the ultrasound focal area. This projection allows focusing through a scattering layer with a localization narrower by a factor 4 in comparison to optical phase conjugation techniques. Moreover, selectively addressing the speckle grains following the excitation profile of the ultrasound is promising.

This work suggests few improvement axes. The use of ultrasonic structured plane wave excitation could benefit from the measurement of the tagging function. These measurements would allow building a matrix of the spectral components of each structuring pattern in the spatial Fourier domain. Inverting this matrix would improve the reconstruction taking in account the full spectral components of the excitation. Moreover, we have seen that using the whole surface of the camera sensor would improve the sensitivity by a factor 16 with respect to the results presented in [chapter 2](#). Nevertheless, this improvement demands to optimize the processing of the image reconstruction by moving to graphical processing units.

The use of ultrasound-scanner probe would be a great improvement for the measurement of the acousto-optic transmission matrix. Indeed transducer array provides a larger numerical aperture, which means it produces more intense ultrasound focus to amplify the tagged-photon signal. In addition the independency of the transducer channels of such probes allows multiple foci, providing the measurement of more complex AOTMs. The use of an ultrasound scanner nevertheless demands to adapt the triggering architecture in order to synchronize the ultrasound excitations with the camera. Then, the acquisition time of the AOTM is currently dominated by the unloading of the camera memory. Many hardware and software alternatives have been tested through the thesis in order to shrink this duration. A camera permitting continuous streaming along triggered recorded should be a significant improvement.

Finally as presented in [chapter 2](#), higher-harmonic modulation has been tested with single-element transducer but not yet using an ultrasound scanner. As the scanner provides arbitrary waveform generation on each of its transducer channel, half-phase random-phase-jumps could be implemented on them to benefit from the sharper point

spread function of the second harmonic.

Appendices

APPENDIX A

Scientific contributions

A.1 Publication

Louis Dutheil, Maïmouna Bocoum, Mathias Fink, Sébastien Popoff, François Ramaz and Jean-Michel Tualle, Fourier transform acousto-optic imaging with off-axis holographic detection, *Applied Optics*, 60(24):7107, 14 August 2021.

A.2 Posters

Louis Dutheil, Maïmouna Bocoum, Sébastien Popoff and François Ramaz, *Acousto-Optic Reflection Matrices*, Imaging in Wave Physics: Multi-Wave and Large Sensor Networks, Cargèse, 23–27 September 2019.

Louis Dutheil, Maïmouna Bocoum, Sébastien Popoff and François Ramaz, *Acousto-Optic Reflection Matrices*, Huitième conférence plénière du GDR Ondes, CentraleSupélec, Gif-sur-Yvette, 28–29 October 2019.

A.3 Oral presentation

Louis Dutheil, Maïmouna Bocoum, Mathias Fink, Sébastien Popoff, François Ramaz, Jean-Michel Tualle, *Imagerie acousto-optique par transformée de Fourier en détection holographique hors-axe*, Congrès annuel de la société française d’optique, Dijon, 5–9 July 2021.

Fourier-Transform Acousto-Optic Imaging with Off-axis Holographic Detection

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Abstract: Acousto-optic (AO) imaging is an in-depth optical imaging technique of highly scattering media. One challenging end-application for this technique is to perform imaging of living biological tissues. Indeed, because it relies on coherent illumination, AO imaging is sensitive to speckle decorrelation occurring on the millisecond timescale. Camera-based detections are well suited for *in vivo* imaging provided their integration time be lower than those decorrelation time scales. In this work, we present Fourier transform acousto-optic imaging combined with off-axis holography which relies on plane-waves and long-duration pulses. We demonstrate, for the first time, a two-dimensional imaging system fully compatible with *in vivo* imaging prerequisites. The method is validated experimentally by performing in-depth imaging inside a multiple scattering sample.

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1. Introduction

Imaging biological tissues at large depths is a challenge inherent to multiple medical applications such as early tumour detection. Indeed, after a few millimetres of propagation, predominance of the multiple scattering regime is detrimental to optical imaging relying on ballistic light in the visible to near-infrared range [1]. Optical coherence tomography [2, 3] is for instance limited to about 100 μm in depth, even when standard wavefront correction [4] is used to compensate for strong aberrations. As a way around, diffuse optical tomography [5] has emerged to infer millimeter-to-centimeter depth optical properties of the medium, based on prior knowledge about scattering properties of the medium [6, 7].

In this context, Photo-Acoustic Imaging (PAI) [8–12] and Acousto-Optic Imaging (AOI) [13, 14] have drawn increasing interest over the past decades. They both provide a direct mapping of optical absorption properties of scattering tissues. In PAI, an intense Q-switched nanosecond laser beam is used to heat optical absorbers inside the medium. As they undergo thermal relaxation, those absorbers emit ultrasound (US) waves in the MHz range which are detected to reconstruct an optically contrasted image. In AOI, the medium is illuminated with a laser of high temporal coherence, while insonified with an auxiliary US pulse in the MHz range. As a result of the AO effect [15], the spectrum of light propagating through the insonified region shows two side-bands which are both shifted from the optical carrier by the US frequency. Those so-called tagged photons which leave the medium are then selectively detected. Due to the local character of the tagging, the absorption properties of the medium can be monitored during US ballistic propagation. Up until now, the development of PAI has been faster than that of AOI, as it relies on incoherent illumination. In fact, for clinical applications, coherent illumination is challenging because of the speckle decorrelation caused by Brownian motion of scatterers, blood circulation, breathing. This decorrelation occurs on time scales in the order or below the millisecond-range [16–19]. In the case of AOI, this sensitivity is highly dependent on the

technique used to detect the tagged photons. In addition, because only a few percent of the light is tagged by the US, weak signal-to-noise ratio (SNR) is another major challenge to address before considering *in vivo* applications.

The most basic scheme to perform AOI is to use temporally-pulsed spatially-focused US for insonification. Using a detection fast enough to follow US pulse propagation, a direct space-to-time mapping allows to reconstruct an in-depth image. Fast detection is for instance performed using photorefractive holography [20] or spectral hole burning [14, 21], both relying on high-sampling-rate photodiode. Robustness against speckle decorrelation makes spectral hole burning promising, but these systems require cryogenic equipment and are not tunable in wavelength.

Camera-based detection [22] of tagged light is an alternative to those detections, provided the camera-integration time remains lower than that of speckle decorrelation. In the particular case of off-axis holography [23], the US frequency-shifted light interferes on the camera with a reference beam of the same frequency. This gives rise to spatial fringes on the detector with a spacing dependent upon the off-axis angle between the two beams. By digitally processing this signal [24, 25], we can retrieve the total number of tagged photons recorded during the camera exposure. Since the camera frames rate ($< \text{kHz}$) is orders of magnitude lower than the sampling rate of a fast photodiode (MHz), it is no longer possible to resolve the propagation of the US pulse which typically progresses at $1.5 \text{ mm } \mu\text{s}^{-1}$. Furthermore, to maximize the number of tagged photons recorded during one camera exposure, temporally-long unfocused US pulses impose as a natural choice because they maximize the insonification volume within the sample. The problem arising with such US wave is a complete loss in spatial resolution, since tagged photons now come from the entire insonification plane.

The problem of recovering spatial resolution using plane and/or continuous wave has been extensively reported in the literature [26, 27]. In the temporal (*i.e.* axial) direction, one solution is to increase the spectral bandwidth of the tagging US, and thereby, that of tagged light. This is achieved for instance by imposing a frequency chirp [28], or random phase jumps [29] on the US carrier frequency. Imposing the same phase modulation on the reference beam when performing off-axis holography, interferences on the camera only build up when the relative delay between the two beams does not exceed the temporal coherence of these modulations. As a result, the tagged photons seem to be emanating from a confined volume along US propagation axis, yielding a recovery of axial resolution. The volume of tagged light however reduces as we increase the spectral bandwidth, which ultimately also degrades the SNR.

To maximize the tagging volume and maintain spatial resolution, Fourier-Transform Acousto-Optic Imaging (FT-AOI) was first introduced using spatially focused US [30, 31]. There, a long US pulse is periodically modulated either in amplitude or phase. When the same modulation is reproduced on the reference arm, tagged photons appear as if generated by several static points periodically distributed along the direction of propagation. Consequently, one acquisition gives access to one component of the image Fourier Transform. In a recent article [32], we have shown that using a transducer array, the same idea could be extended to fetch the two-dimension Fourier components of the image. However, this work relied on photorefractive-based holographic detection, and therefore sensitive to speckle decorrelation. In this paper we present, for the first time, FT-AOI combined with off-axis heterodyne holographic detection.

2. Theory

The theoretical framework of the method is described in [30]. The present section focuses on true implementation of the method using a transducer array [32]. In FT-AOI, the pressure field is periodically structured along the x and z -axis, corresponding respectively to the direction along the transducer array and the direction of acoustic propagation. Reconstructing an image requires to perform multiple acquisitions so as to fetch all of its Fourier components. For each acquisition,

the structuring frequencies ν_x along x-axis is an increasing harmonic of fundamental frequencies $\nu_x^0 = L^{-1}$ where L is the width of the US probe, such that $\nu_x = m \nu_x^0$ with m integer. The modulation along z is done by imposing a periodic temporal emission on each transducer with a relative phase imposed by the periodicity set along x . At any given time during propagation, the field thereby appears spatially modulated along z at a frequency $\nu_z = n \nu_z^0$, where n is an integer corresponding to the harmonic order and ν_z^0 the fundamental modulation frequency. By definition, we have that $\nu_z^0 = (c_s T_0)^{-1}$ with T_0 the user-defined fundamental temporal period and c_s the sound velocity inside the medium. In addition, by controlling the emission time on all transducers simultaneously, we can vary the relative phase ϕ of the structuring pattern. We note $P_{m,n}^\phi(x, t)$ the pressure field emitted by the transducer array for the (m, n) -harmonic pattern and a phase ϕ at $z = 0$ and for $0 \leq x \leq L$:

$$P_{m,n}^\phi(x, t) = P_0 h_{m,n}^\phi(t, x) \cos \omega_{us} t. \quad (1)$$

where $h_{m,n}^\phi$ stands for the adimensional amplitude of the modulation and P_0 for the nominal pressure. At each position x , $h_{m,n}^\phi(\bullet, x)$ is a T_0/n -periodic function. On one period, it equals to 1 when $0 \leq t < T_0/2n$ and 0 otherwise, such that $(h_{m,n}^\phi)^2 = h_{m,n}^\phi$. By construction, we also impose that function $h_{m,n}^\phi(t, \bullet)$ be L/m -periodic along x . This function is represented on Fig.1 for different values of n , m and ϕ . By neglecting diffraction effects upon propagation, the pressure

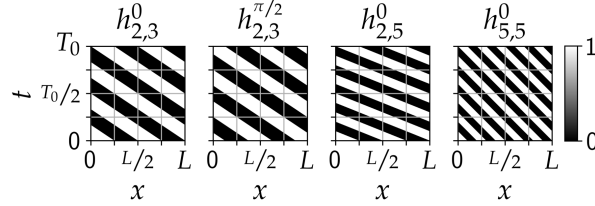


Fig. 1. Examples of the amplitude of modulation $h_{m,n}^\phi$

field at time t following the emission writes:

$$P_{m,n}^\phi(x, z, t) \equiv A_{m,n}^\phi(x, z, t) \cos [\omega_{us}(t - c_s^{-1} z)], \quad (2)$$

$$A_{m,n}^\phi(x, z, t) = P_0 h_{m,n}^\phi(t - c_s^{-1} z, x), \quad (3)$$

with ω_{us} the US carrier pulsation and P_0 the nominal pressure. As US propagate, some photons are frequency-shifted from the incident optical carrier ω_L at $\omega_L \pm \omega_{us}$ with a phase and amplitude inherited from $A_{m,n}^\phi$. In the holographic digital detection scheme, the collected field E interferes with a reference field E_{ref} centered at $\omega_L + \omega_{us}$. The resulting image $I_{m,n}^\phi(\eta, \xi)$ obtained over an integration time τ_e writes:

$$I_{m,n}^\phi(\eta, \xi) = \epsilon_0 \int_{\tau_e} dt |E_{ref}(\eta, \xi, t) + E(\eta, \xi, t)|^2 \quad (4)$$

where (η, ξ) are the pixels coordinates on the camera sensor. By imposing that E_{ref} also be T_0/n periodic in amplitude, constructive interference over the integration time τ_e will only build up for tagged light coming from specific locations in the interaction plane. These locations are defined by our so called "tagging function". The temporal expression of the reference field incident on the camera writes:

$$E_{ref}(\eta, \xi, t) = E_0 \sin^2 \left(\pi \frac{n}{T_0} t \right) e^{-i \frac{\omega_L + \omega_{us}}{c} (\alpha_\eta \eta + \alpha_\xi \xi)} e^{i(\omega_L + \omega_{us})t}, \quad (5)$$

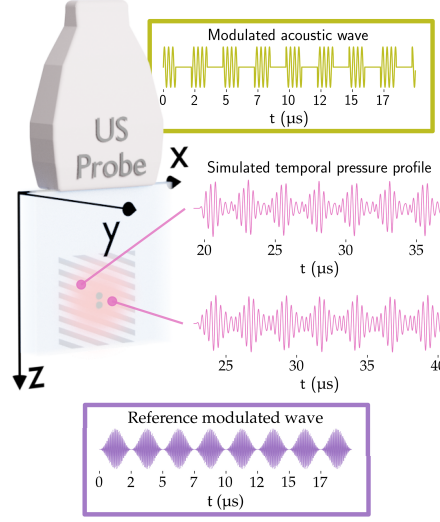


Fig. 2. Principle of the method. The two-dimensional probe sends a temporally modulated acoustic beam (top caption) specified by $h_{m,n}^\phi$. The acoustic field which propagates inside the scattering medium is represented at two different arbitrary positions inside the sample. The tagging function will result in the cross-correlation of this local field with the reference wave (lower caption).

where c is the velocity of light, E_0 is the field amplitude, and α_η and α_ξ the off-axis angles.

Following the basic principle of off-axis holography, the recorded image is numerically filtered in the Fourier domain so as to only keep the tilted component. This operation leads to the following hologram resulting from the interferences between the scattered light and the reference beam:

$$H_{m,n}^\phi(\eta, \xi) = \epsilon_0 e^{i \frac{\omega_L + \omega_{us}}{c} (\alpha_\eta \eta + \alpha_\xi \xi)} \int_{\tau_e} dt E_0^* E(\eta, \xi, t) \sin^2 \left(\pi \frac{n}{T_0} t \right) e^{-i(\omega_L + \omega_{us})t} \quad (6)$$

The averaged square modulus of this hologram on all of the pixels, $s_{m,n}^\phi \equiv \langle |H_{m,n}^\phi|^2 \rangle$, is proportional to the average intensity of the tagged photons selectively detected in the US interaction plane [30]. We write it in the following form:

$$s_{m,n}^\phi \propto \int_{x,z} dx dz I(x, z) \left[M_{m,n}^\phi(x, z) \right]^2. \quad (7)$$

where $I(x, z)$ is the image we wish to reconstruct, which depends on the local average intensity of the diffuse light, on the tagging efficiency, and on the propagation from the medium to the camera [30]. $M_{m,n}^\phi(x, z)$ is the tagging function which ensures spatial selectivity. As illustrated in Fig 2, the acoustic field modulation pattern has an arrival phase which varies along x and z . As a result, tagged photons will only be fully correlated with the reference wave at specific positions (x, z) . The expression of tagged photons is easily derived from Eq 2, such that the tagging function becomes the following cross-correlation [30]:

$$\begin{aligned} M_{m,n}^\phi(x, z) &= \frac{P_0}{\tau_e} \int_{\tau_e} dt \sin^2 \left(\pi \frac{n}{T_0} t \right) h_{m,n}^\phi(t - c_s^{-1} z, x) \\ &= \frac{P_0}{2} \left[1 + \frac{1}{\pi} \cos \left(2\pi m v_x^0 x + 2\pi n v_z^0 z + \phi \right) \right] \end{aligned} \quad (8)$$

Detailed derivation of Eq 8 is performed in Supplement 1. This expression shows that the sinusoidal modulation applied on the reference beam leads to periodically distributed tagged photons both in x and z -axis. By combining measurements obtained using four different phase shifts, namely $\phi \in \{0, \pi/2, \pi, 3\pi/2\}$, we access the complex Fourier components $\tilde{I}$ of the acousto-optic image:

$$s_{m,n}^{3\pi/2} - s_{m,n}^{\pi/2} + i \left(s_{m,n}^{\pi} - s_{m,n}^0 \right) \propto \tilde{I} \left(m v_x^0, n v_z^0 \right), \quad (9)$$

Image $I(x, z)$ is thus simply reconstructed by performing an inverse Fourier Transform (iFT) of $\tilde{I}$. In our model, we however recall that diffraction is neglected (Eqs 2 and 8), an hypothesis which might not always verify experimentally. Thus, to investigate potential effect of diffraction, we have performed a simulation of the actual pressure emitted by the probe we used in our later described experiment. Simulations were performed using Field II open-source software as already described in [33]. A simulation grid is placed in the US plane, 35mm below the probe emission surface with $-15 \leq x \leq 15$ and $20 \leq z \leq 50$. On each point of a simulation grid, we have calculated the tagging function using the simulated pressure and the expression of our theoretical optical reference field. As a result, we conclude that diffraction effects can indeed be neglected for all n (tested up to 20). As m value increases however, second harmonic artefacts start to appear in the tagging function, together with π -phase shifts of the sin wave. This effect is to be expected as a result of Talbot near-field diffraction effect and will be fully explained in future work. Example of resulting $M_{m,n}^{\phi}$ functions are shown Fig.3 for $m = 3, 5, 6$ and $n = 8$. In our simulated experimental conditions, diffraction artefacts start to appear typically for $|m| \geq 6$ with a noticeable phase shift at ~ 38 mm. For this reason, we constrained our experiment to $|m| < 6$ so as to remain with the scope of validity of our model.

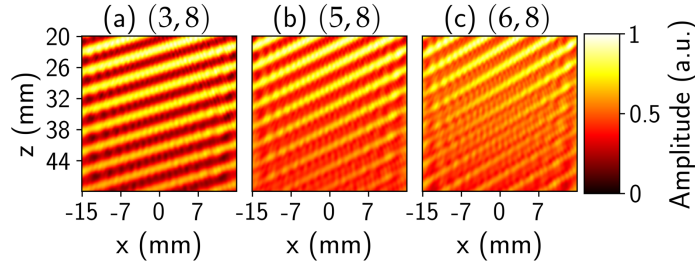


Fig. 3. Simulated tagging functions for $m = 3$ (a), 5 (b) and 6 (c), $n = 8$ on a $20 \leq z \leq 50$ mm and $-15 \leq x \leq 15$ mm simulation grid.

3. Experiment

The sample is an hydrogel mimicking both the optical and mechanical properties of soft biological tissues [34]. The dimensions of the hydrogel along x , y and z axes are respectively $5 \times 1 \times 5$ cm³. The 50 kPa Young modulus is set by the 2 % concentration of Agar [35] whereas the concentration of Intralipid 10 % is adjusted to obtain a reduced scattering coefficient $\mu'_s = 1$ cm⁻¹ (transport mean free path $l^* = 1$ cm) [36]. Two ink inclusions are inserted at a 5 mm depth. The dimensions of their cylindrical shapes are 4 mm in height and 2 mm of diameter. Their centers are separated by 4 mm. A photograph showing the two inclusions taken during the molding is shown on Fig.4. The sample is illuminated with a single-longitudinal mode laser centered at 780 nm. After the tapered amplifier (MOPA, Sacher Lasertechnik GmbH), the incident power is about 370 mW for a beam diameter of 13 mm. A transmitted photons flux of 3.6 μ W is collected by the CMOS camera (Ximea xiB-64) located 34 cm behind the sample output surface. This distance corresponds to a

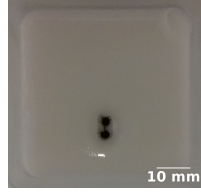


Fig. 4. Photograph of the two ink inclusions taken above the gel cast. The two inclusions stands at 35 mm from the top surface the gel. This distance corresponds to the elevation focus of the probe.

trade-off between proper sampling of the interference fringes and a maximization of the optical collection. A scheme of the experimental setup is presented on Fig.5.

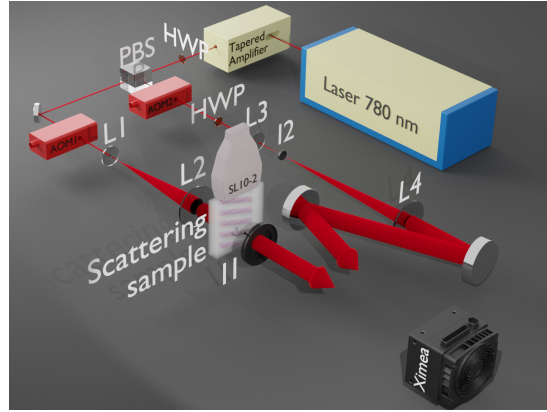


Fig. 5. Experimental setup. Light emitted by a laser diode at 780 nm is split between the probe beam and the reference arm with a half-wave plate (HWP) followed by a polarizing beam-splitter (PBS). Both arms are then modulated in frequency by two acousto-optic modulators (AOM#) around 80 MHz with a relative difference of 3 MHz corresponding to the carrier frequency of the acoustic waves emitted by the transducer array (SL10-2 – Aixplorer SL10-2 US probe, SuperSonic Imagine, Aix-en-Provence, France) through the scattering hydrogel. Lenses (L#) and irises (I#) widen and filter the beams before they meet the camera (Ximea) sensor.

The structured US plane waves are emitted with a 192-element linear transducer array (Aixplorer SL10-2 US probe, SuperSonic Imagine, Aix-en-Provence, France) with a fixed focal elevation of 35 mm. The excitation voltage of the transducer is set to 15 V such that the nominal pressure of the US wave was measured to be $P_0 \approx 300$ kPa. Such a pressure corresponds to a US peak intensity of $\approx 3 \text{ W cm}^{-2}$. Accounting for the acquisition repetition rate, the average acoustic intensity is $\approx 150 \text{ mW cm}^{-2}$, far below biomedical norms of 720 mW cm^{-2} [37]. We set the fundamental temporal window of our periodic pattern to $T_0 = 20 \mu\text{s}$. The pattern is then repeated five times so as to match the exposure time of the camera $\tau_e = 100 \mu\text{s}$. The camera is triggered at a 300 Hz repetition rate and each component (given n, m, ϕ value) is measured with a single-shot image. Here, we only use a 1024×1024 pixel area of the 16 Mpixel chip, to prevent frame dropping which occurs when our frame grabber gets filled up. In fact, our image processing performed using Labview software runs on multi-core CPU unit, but remains too slow to prevent this buffer saturation at full chip capacity. A full frame acquisition would improve the SNR as the sensor surface is increased but would require Graphics Processing Unit (GPU)

for the reconstruction process. The total acquisition time to image a complete field of view of $30 \times 30 \text{ mm}^2$ is 1.5 s.

We image the two ink inclusions hidden within hydrogels of two different reduced scattering coefficient 1 cm^{-1} and 10 cm^{-1} . The image corresponding to the $\mu'_s = 1 \text{ cm}^{-1}$ hydrogel featured on Fig.6 shows two dark spots in the halo of the tagged beam. with a diameter of 2 mm, and separated by 4 mm, as expected. This image was reconstructed from the 110 complex Fourier components, $m \in \llbracket -5, 5 \rrbracket$, $n \in \llbracket 1, 10 \rrbracket$ which modulus is shown in Fig.6(c). The resolution of FT-AOI is given by the spatial period of the higher harmonics used in the structuring pattern $h_{m,n}$ with the wavelength of the US carrier as the ultimate limit. As the maximum values for m and n are respectively $m = 5$ and $n = 10$, the resolutions along x and z are respectively $(m \nu_x^0)^{-1} = 7.7 \text{ mm}$ and $(n \nu_z^0)^{-1} = 3 \text{ mm}$, which discriminates our two inclusions.

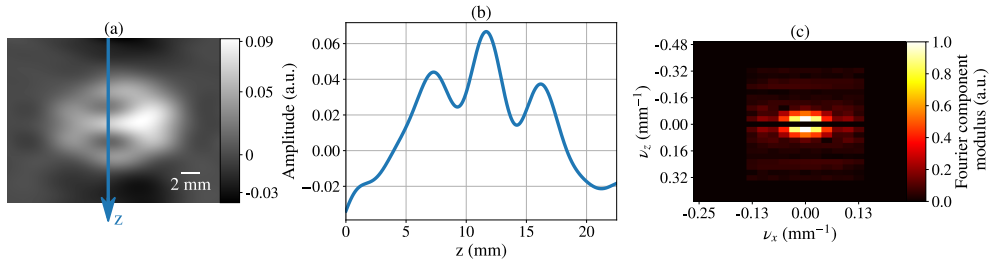


Fig. 6. FT-AOI of two ink inclusions buried within a 1 cm-thick scattering hydrogel with a reduced scattering coefficient of 1 cm^{-1} . (a) The image is obtained by gathering the 110 experimentally measured Fourier components for $-5 \leq m \leq 5$ and $1 \leq n \leq 10$. The profile (b) is drawn following the blue line on the acousto-optic image crossing the two dark spots. (c) Modulus of the 4-phase acquired Fourier components.

To outline time-efficiency of FT-AOI, we draw a comparison with AO imaging in real space as for instance performed using spatially-focused US with random phase jumps modulation [29, 38]. In this respect, let's assume such an image be sampled every $\sim 2 \text{ mm}$ over a $30 \times 30 \text{ mm}^2$ field of view. This corresponds to $15 \times 15 = 225$ points to acquire. For each sampled point, the AO detected signal in experimental conditions similar as in present work is nearly one to two orders of magnitude lower [38] than for FT-AOI. In fact, because the tagging volume is reduced to few cubic millimeters, it requires a proportional increase in the number of averaging. As a result, the acquisition time for an image acquired in real space is on the order of a minute. In FT-AOI where acquisition is performed in reciprocal space, the tagging volume is significantly increased such that averaging is no longer required. As a result, the method is typically one order of magnitude more time efficient.

For the stronger-diffusive hydrogel $\mu_s = 10 \text{ cm}^{-1}$, the transmitted intensity through the sample is 6 times weaker than in previous case. This decreases the SNR to ≈ 1.6 . Fourier components are here again fetched without averaging, and modulus of the noisy Fourier components are shown in Fig.7 (c). The two inclusions are still discriminated. The noise however affects the image reconstruction as we see on Fig.7 (a-b). We observe a phase offset between high and low frequency components such that several inclusions of different sizes seem to appear, which shows the limitation of the method at low SNR. We nevertheless emphasize that this result is obtained without averaging, thereby demonstrating the robustness of our imaging strategy.

In conclusion, we have shown that the FT-AOI combined with an off-axis detection can achieve averaging-free imaging over a $3 \times 3 \text{ cm}^2$ field-of-view with mm resolution. This camera-based detection integrates the energy of multiple speckle grains impinging on the detector. A reference beam is used to extract and demodulate the tagged signal, and as a result of autocorrelation, only photons allowed by the tagging function are detected. This allows to perform imaging in the

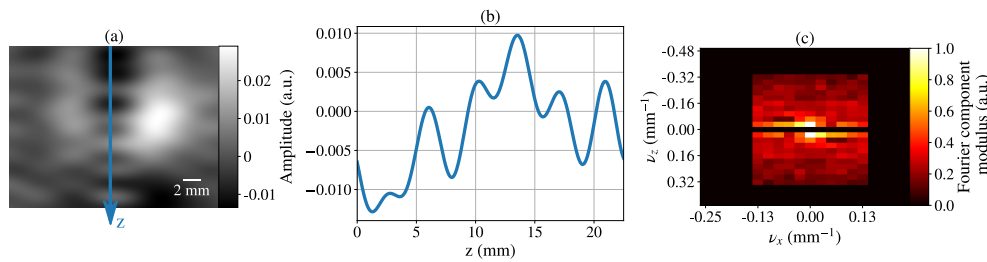


Fig. 7. FT-AO images of two ink inclusions embedded in 1 cm-thick scattering hydrogel with a reduced scattering coefficient of 10 cm^{-1} . (a) The image is obtained by gathering the 110 experimentally measured Fourier components for $-5 \leq m \leq 5$ and $1 \leq n \leq 10$. The profile (b) is drawn following the blue line on the acousto-optic image crossing the two dark spots. (c) Modulus of the 4-phase acquired Fourier components.

Fourier domain of the object while still tagging a large volume inside the sample. The camera integration time is kept below a millisecond such as to be insensitive to speckle decorrelation. In addition, the overall acquisition time of an image was 1.5 s, making this method adapted for *in vivo* monitoring.

This short acquisition time is only possible because of the large volume of tagged photon inherent to the method, even with limited effective collection on the camera and low pressure peak powers. We plan to further increase the image SNR by using the full camera chip along with GPU stream analysis. In this article, image reconstruction was performed using a straightforward two-dimensional inverse Fourier transform. In the future, we plan to investigate other reconstruction strategies, in particular one which can account for diffraction effects visible on the tagging function. This can be done for instance by measuring an acousto-optic tagging matrix, thereby opening the way to wide variety of tagging strategy for optimal imaging.

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Disclosures. The authors declare no conflicts of interest.

Data availability. Data underlying the results presented in this paper are not publicly available at this time but may be obtained from the authors upon reasonable request.

Supplemental document. See Supplement 1 for supporting content.

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ACOUSTO-OPTIC REFLECTION MATRICES

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Motivation

Acousto-optic imaging allows to probe locally **optical contrast** of biological media at **depths** where multiple scattering of light by inhomogeneities limits microscopy. However **resolution** of imaging is **limited by the wavelength of ultrasounds** which is many order of magnitude larger than resolution in optical microscopy. Using techniques of wavefront shaping, we purpose to mesure **acousto-optic reflection matrices (AORM)**. AORM links the incoming optic field to the backscattered field of ultrasonically tagged photons in scattering media. The acquisition of multiple matrices moving the ultrasonic source will allow us to get information on spatial variations smaller than the acoustic wavelength. Hence it improves the imaging resolution bringing it closer to the limit of **optical diffraction**.

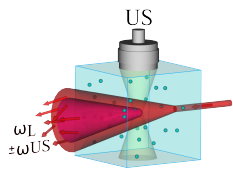
Acousto-optic effect

Basics

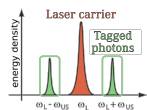
Optical path modulation:

- * Motion of scatterer (Doppler effect)
- * Variation of pressure
- * Doppler effect

Modulation volume *i.e.* volume of optical sources $\propto \lambda_{US}^3 \sim mm^3$



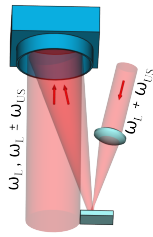
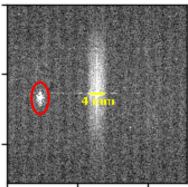
Frequency shift



- * $f_{out} = f_{in} \pm f_{US}$
- * $f_{in} \sim 10^{14} Hz$
- * $f_{US} \sim 10^7 Hz$

Select tagged photons – Off-axis holography

- * A reference beam at the tagged-photon frequency, angle $\theta \sim 1^\circ$.
- * Amplification and shift the contribution of **tagged photons** in spatial Fourier domain.



Hologram intensity:

$$H = \|E_R + E_S\|^2 = \|E_R\|^2 + \|E_S\|^2 + E_R E_S^* + E_R^* E_S = I_R + I_S + E_R E_S^* + E_R^* E_S$$

Spatial sectioning

Random phase shift modulation

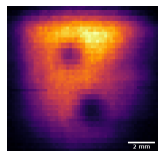
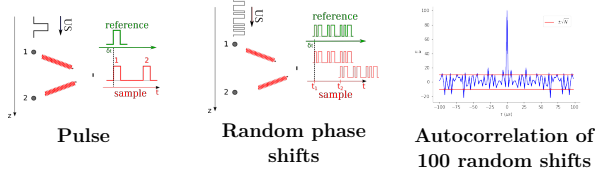
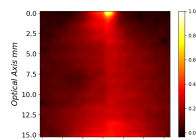


Image of 2 inclusions



Acousto-optic signal across 10%-Intralipid® – agar gel ($l^* = 1mm$) – 2cm-thick

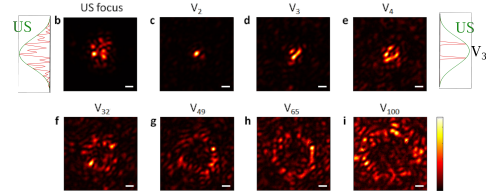
Focusing beyond the acoustic limit

The acousto-optic transmission matrix (AOTM) gives the relation between the illumination input (shaped by an SLM) and the AO tagged signals (recorded by the CCD camera) through a scattering medium:

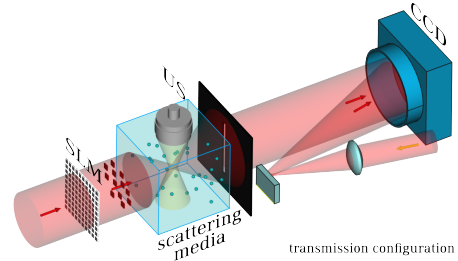
$$E^{out}(f_L + f_{US}) = T E^{in}(f_L)$$

We can exploit the AOTM to maximise the transmitted signal.

The first singular vectors of $TT^\dagger$ confines the input illumination into the US focus¹.



Focusing in a volume sample

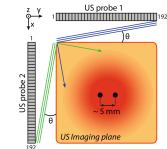


Next?

Structured ultrasound modulation²

Use plane waves

- * Image a larger volume
- * Scan a volume sample quicker
- * Better SNR than US in standard focused regime



Move to reflection configuration

Observe the illumination backscattered by the sample. In most imaging condition only one side of biological tissue is accessible. $\Rightarrow$ **Weaker signal**

Theoretical application

Create artificial acousto-optic sources inside the sample:

- * Probe statistical properties, information between targets
- * Impact of mesoscopic correlations



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APPENDIX B

Developed derivations in FT-AOI

THIS section develops and gives more details about the Fourier transform acousto-optic imaging of an insonified plane, described in [subsection 2.4.1](#). According to the definition given in the main body of this article, the complex correlation function $M_{m,n}^\phi$ exhibits the selection operated between the two modulations during the exposure time τ_c of the camera. Here we fully develop its analytical derivation.

$$M_{m,n}^\phi(x, z) = \frac{1}{\tau_c} \int_{\tau_c} dt \sin^2 \left(\pi \frac{n}{T_0} t \right) A(x, z, t). \quad (\text{B.1})$$

As presented in the article, the pressure field in propagation accross the plane (x, z) , x transverse coordinate along the axis of the probe, z along the US propagation axis, has the expression:

$$A(x, z, t) = P_0 h_{m,n}^\phi(t - c_s^{-1} z, x). \quad (\text{B.2})$$

Replacing [Equation B.2](#) in [Equation B.1](#) gives:

$$M_{m,n}^\phi(x, z) = \frac{P_0}{\tau_c} \int_{\tau_c} dt \sin^2 \left(\pi \frac{n}{T_0} t \right) h_{m,n}^\phi \left(t - c_s^{-1} z, x \right). \quad (\text{B.3})$$

The analytical expression of $h_{m,n}^\phi$ is:

$$h_{m,n}^\phi(t, x) = \sum_k \Pi_n \left(t - \frac{m \nu_x^0}{n \nu_z^0} \frac{x}{c_s} - \frac{k T_0}{n} + \frac{\phi T_0}{2\pi n} \right), \quad (\text{B.4})$$

where Π_n is a symmetric square of period T_0/n :

$$\Pi_n(t) = \begin{cases} 1 & \text{if } 0 \leq t < T_0/2n \\ 0 & \text{otherwise} \end{cases}. \quad (\text{B.5})$$

With this definition of Π_n , $h_{m,n}^\phi$ is a binary function built up with n cycles of period T_0/n . We draw it on [Figure B.1](#). We use the trigonometric identity:

$$\sin^2 \left(\pi \frac{n}{T_0} t \right) = \frac{1}{2} \left[1 - \cos \left(2\pi \frac{n}{T_0} t \right) \right]. \quad (\text{B.6})$$

Experimentally we can choose the integration time of the camera independently from the temporal length of the sequence. In order to we choose commensurable quantities such as:

$$\tau_c = N T_0 \quad \nu_x^0 = L^{-1} \quad \nu_z^0 = (c_s T_0)^{-1}, \quad (\text{B.7})$$

with N an integer, L the length of the probe. For the image in section we have chosen $N = 5$ for $T_0 = 20 \mu\text{s}$ and $\tau_c = 100 \mu\text{s}$. The commensurability of τ_c and T_0 allows to have the same total insonified time during the measurement for any couple of parameters m and n . This quantity corresponds to the integration of binary function $h_{m,n}^\phi$ represented on [Figure B.1](#).

$$\forall(m, n) \quad \forall x \in [0, L] \quad \int_{\tau_c} h_{m,n}^\phi(t, x) dt = \frac{N T_0}{2}. \quad (\text{B.8})$$

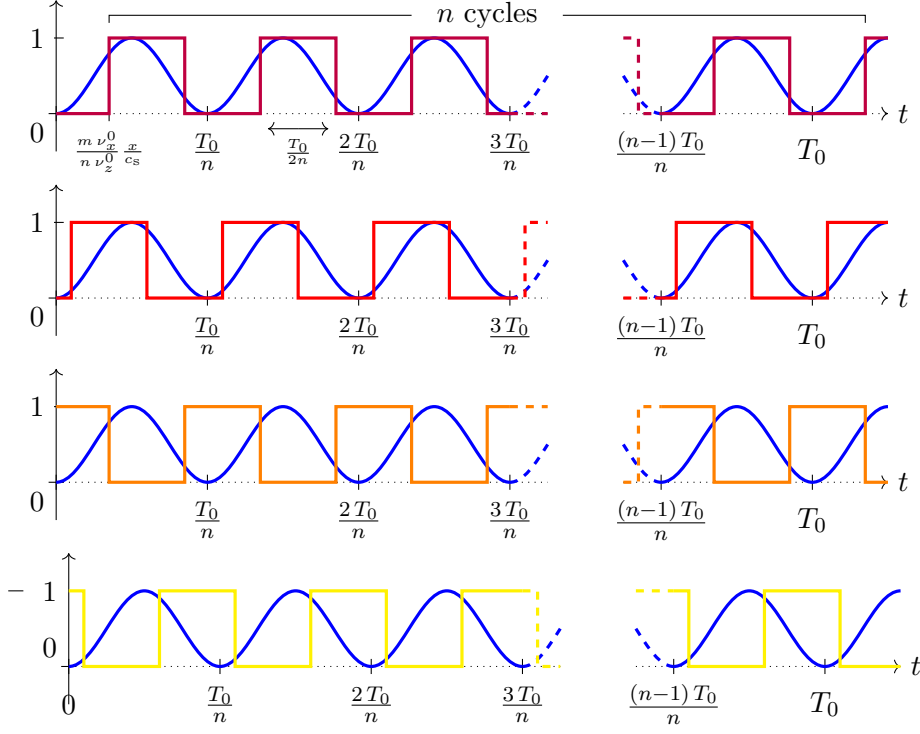


Figure B.1 – $\sin^2(\pi n t/T_0)$ in blue and $h_{m,n}^\phi(t, x)$ for an arbitrarily chosen position x and for $\phi \in \{0, \pi/2, \pi, 3\pi/2\}$ in respectively $\{\blacksquare, \blacksquare, \blacksquare, \blacksquare\}$.

Using Equation B.6 and Equation B.8 in Equation B.3, we have:

$$M_{m,n}^\phi(x, z) = \frac{P_0}{2NT_0} \int_{NT_0} dt \left[1 - \cos\left(2\pi \frac{n}{T_0} t\right) \right] h_{m,n}^\phi\left(t - c_s^{-1}z, x\right), \quad (\text{B.9})$$

$$= \frac{P_0}{2} \left[\frac{1}{2} - \frac{1}{NT_0} \int_{NT_0} dt \cos\left(2\pi \frac{n}{T_0} t\right) h_{m,n}^\phi\left(t - c_s^{-1}z, x\right) \right]. \quad (\text{B.10})$$

As $h_{m,n}^\phi$ is a binary T_0/n -periodic function, we can split the integral into $n \times N$ small narrower integrals (see Figure B.1 for a graphical example):

$$M_{m,n}^\phi(x, z) = \frac{P_0}{2} \left[\frac{1}{2} - \frac{n}{T_0} \int_{\frac{m\nu_z^0}{n\nu_z^0} \frac{x}{c_s} + \frac{z}{c_s} - \phi + \frac{T_0}{2n}}^{\frac{m\nu_x^0}{n\nu_z^0} \frac{x}{c_s} + \frac{z}{c_s} - \phi + \frac{T_0}{2n}} dt \cos\left(2\pi \frac{n}{T_0} t\right) \right], \quad (\text{B.11})$$

$$= \frac{P_0}{2} \left[\frac{1}{2} - \frac{n}{T_0} \int_{\frac{m\nu_z^0}{n\nu_z^0} \frac{x}{c_s} + \frac{z}{c_s} - \phi}^{\frac{m\nu_x^0}{n\nu_z^0} \frac{x}{c_s} + \frac{z}{c_s} - \phi + \frac{T_0}{2n}} dt \cos\left(2\pi \frac{n}{T_0} t\right) \right], \quad (\text{B.12})$$

$$M_{m,n}^\phi(x, z) = \frac{P_0}{2} \left(\frac{1}{2} - \frac{1}{2\pi} \left\{ \sin \left[2\pi \left(m \nu_x^0 x + n \nu_z^0 z \right) - \phi + \pi \right] \right. \right. \\ \left. \left. - \sin \left[2\pi \left(m \nu_x^0 x + n \nu_z^0 z \right) - \phi \right] \right\} \right), \quad (\text{B.13})$$

$$= \frac{P_0}{2} \left[\frac{1}{2} + \frac{1}{\pi} \sin(2\pi(m \nu_x^0 x + n \nu_z^0 z) - \phi) \right]. \quad (\text{B.14})$$

This operation leads to the following hologram resulting from the interferences between the scattered light and the reference beam:

$$H_{m,n}^\phi(\eta, \xi) \propto e^{i \frac{\omega_L + \omega_{\text{US}}}{c} (\alpha_\eta \eta + \alpha_\xi \xi)} \int_{\tau_c} dt E_0^* E(\eta, \xi, t) \sin^2 \left(\pi \frac{n}{T_0} t \right) e^{-i(\omega_L + \omega_{\text{US}})t}. \quad (\text{B.15})$$

The averaged square modulus of this hologram, $s_{m,n}^\phi \equiv \langle |H_{m,n}^\phi|^2 \rangle$, is proportional to the average intensity of the tagged photons selectively detected in the US plane (Barjean et al., 2016). We write it in the following form:

$$s_{m,n}^\phi \propto \int_{x,z} dx dz I(x, z) |M(x, z)|^2. \quad (\text{B.16})$$

Although we define $I(x, z)$ as the acousto-optic image within the medium, $I(x, z)$ is a real function. We define $\tilde{I}$ by the 2D-Fourier transform of I .

$$s_{m,n}^\phi \propto \int_{x,z} dx dz I(x, z) \left\{ \frac{1}{2} + \frac{1}{\pi} \sin \left[2\pi \left(m \nu_x^0 x + n \nu_z^0 z \right) - \phi \right] \right\}^2, \quad (\text{B.17})$$

$$\propto \int_{x,z} dx dz I(x, z) \left\{ \frac{1}{4} + \frac{1}{2\pi^2} + \frac{1}{\pi} \sin \left[2\pi \left(m \nu_x^0 x + n \nu_z^0 z \right) - \phi \right] \right. \\ \left. - \frac{1}{2\pi^2} \cos \left[4\pi \left(m \nu_x^0 x + n \nu_z^0 z \right) - 2\phi \right] \right\}, \quad (\text{B.18})$$

$$\propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} dx dz I(x, z) \\ + \frac{1}{2\pi i} \left[-\tilde{I} \left(m \nu_x^0, n \nu_z^0 \right) e^{i\phi} + \tilde{I} \left(-m \nu_x^0, -n \nu_z^0 \right) e^{-i\phi} \right] \\ - \frac{1}{4\pi^2} \left[\tilde{I} \left(2m \nu_x^0, 2n \nu_z^0 \right) e^{2i\phi} + \tilde{I} \left(-2m \nu_x^0, -2n \nu_z^0 \right) e^{-2i\phi} \right]. \quad (\text{B.19})$$

As I is a real function:

$$\tilde{I} \left(m \nu_x^0, n \nu_z^0 \right) = \tilde{I}^* \left(-m \nu_x^0, -n \nu_z^0 \right), \quad (\text{B.20})$$

and

$$\tilde{I} \left(2m \nu_x^0, 2n \nu_z^0 \right) = \tilde{I}^* \left(-2m \nu_x^0, -2n \nu_z^0 \right), \quad (\text{B.21})$$

so

$$s_{m,n}^\phi \propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} dx dz I(x, z) - \frac{1}{\pi} \Im \left[\tilde{I} \left(m \nu_x^0, n \nu_z^0 \right) e^{i\phi} \right] \\ - \frac{1}{2\pi^2} \Re \left[\tilde{I} \left(2m \nu_x^0, 2n \nu_z^0 \right) e^{2i\phi} \right]. \quad (\text{B.22})$$

Measurements for various phase shifts, $\phi \in \{0, \pi/2, \pi, 3\pi/2\}$, give access to the real and the imaginary parts:

$$s_{m,n}^0 \propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} I(x, z) - \frac{1}{\pi} \Im \left[\tilde{I}(m\nu_x^0, n\nu_z^0) \right] - \frac{1}{2\pi^2} \Re \left[\tilde{I}(2m\nu_x^0, 2n\nu_z^0) \right], \quad (\text{B.23})$$

$$s_{m,n}^{\pi/2} \propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} I(x, z) - \frac{1}{\pi} \Re \left[\tilde{I}(m\nu_x^0, n\nu_z^0) \right] + \frac{1}{2\pi^2} \Re \left[\tilde{I}(2m\nu_x^0, 2n\nu_z^0) \right], \quad (\text{B.24})$$

$$s_{m,n}^{\pi} \propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} I(x, z) + \frac{1}{\pi} \Im \left[\tilde{I}(m\nu_x^0, n\nu_z^0) \right] - \frac{1}{2\pi^2} \Re \left[\tilde{I}(2m\nu_x^0, 2n\nu_z^0) \right], \quad (\text{B.25})$$

$$s_{m,n}^{3\pi/2} \propto \left(\frac{1}{4} + \frac{1}{2\pi^2} \right) \int_{x,z} I(x, z) + \frac{1}{\pi} \Re \left[\tilde{I}(m\nu_x^0, n\nu_z^0) \right] + \frac{1}{2\pi^2} \Re \left[\tilde{I}(2m\nu_x^0, 2n\nu_z^0) \right]. \quad (\text{B.26})$$

The linear combination yields to the complex Fourier component:

$$s_{m,n}^{3\pi/2} - s_{m,n}^{\pi/2} + i(s_{m,n}^{\pi} - s_{m,n}^0) \propto \tilde{I}(m\nu_x^0, n\nu_z^0). \quad (\text{B.27})$$

APPENDIX C

Experimental electrical connections and timing tables

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C.1 Fourier transform imaging experimental setup

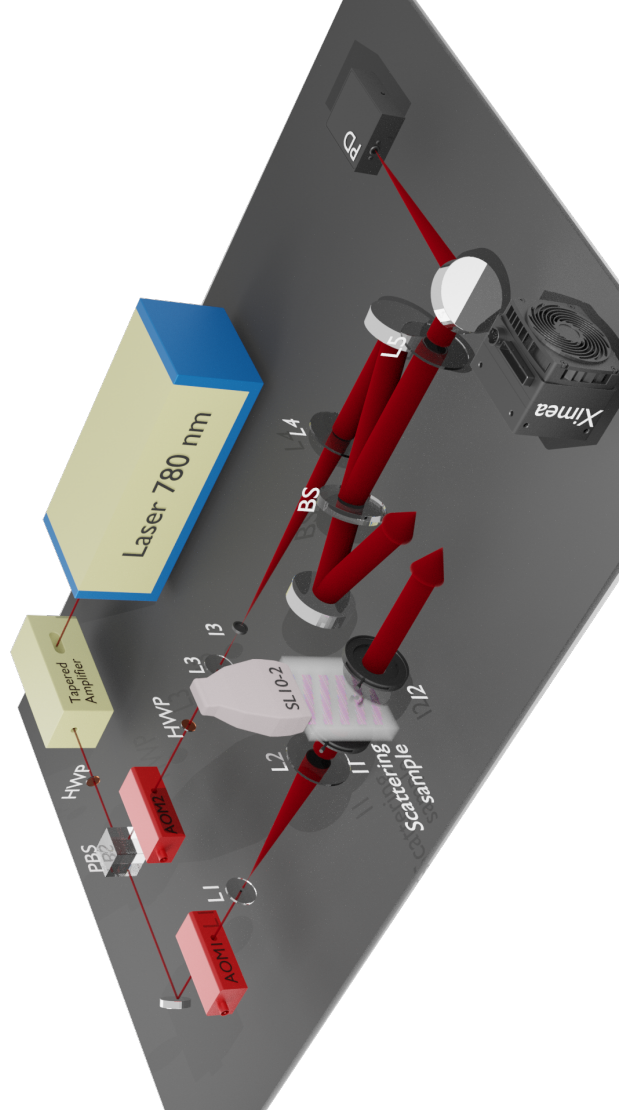


Figure C.1 – Experimental setup used for the FT-AOI, light emitted by a laser diode at 780 nm is split between the probe beam and the reference arm with a half-wave plate (HWP) followed by a polarizing beam-splitter (PBS). Both arms are then modulated in frequency by two acousto-optic modulators (AOM#) around 80 MHz with a relative difference of 3 MHz corresponding to the carrier frequency of the acoustic waves emitted by the transducer array (SL10-2 – Aixplorer SL10-2 US probe, SuperSonic Imagine, Aix-en-Provence, France) through the scattering hydrogel. Lenses (L#) and irises (I#) widen and filter the beams before they meet the camera (Ximea) sensor.

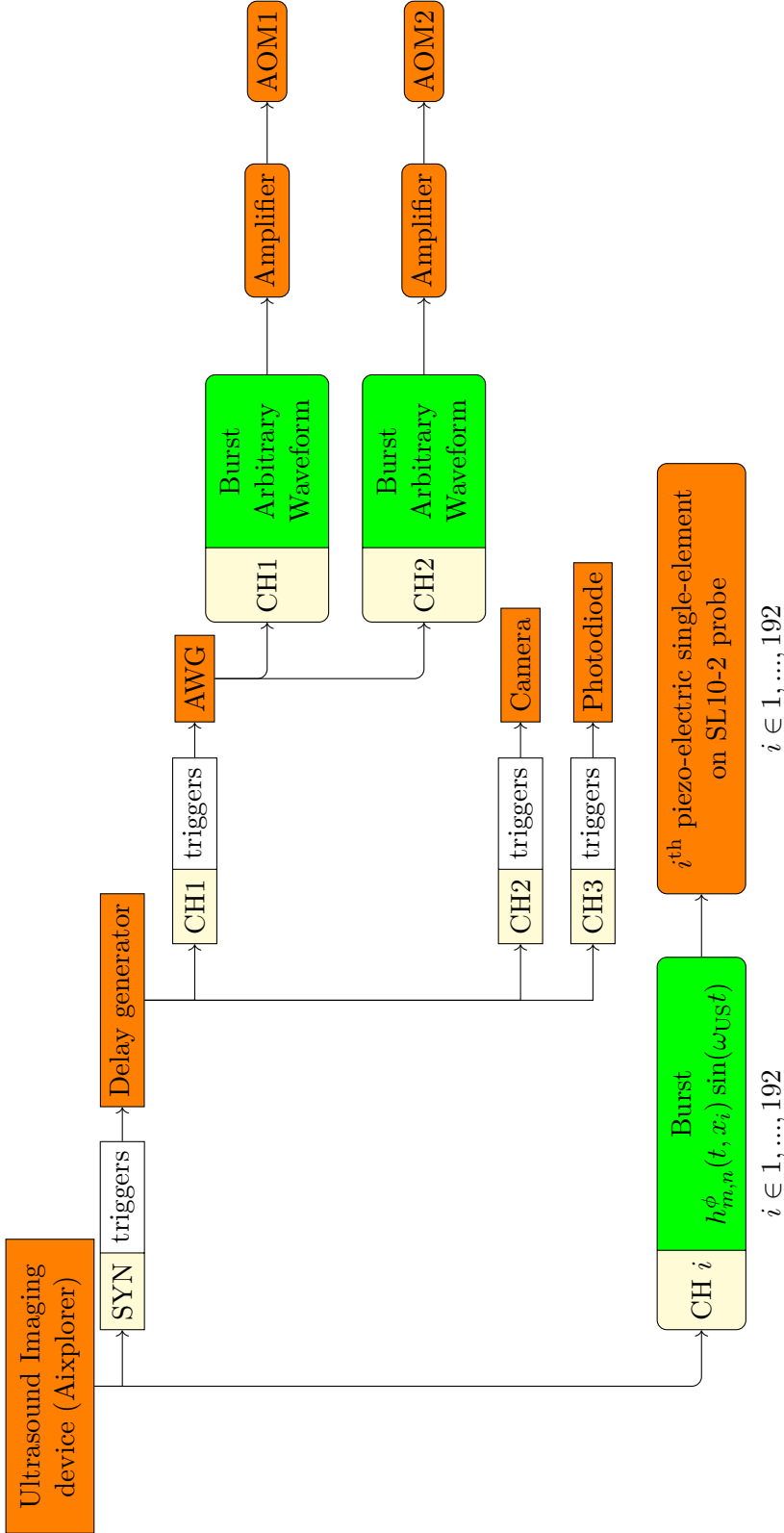


Figure C.2 – Electric connection schematic of the devices used on the FT-AOI experiments. Aixplorer: ultrasound device developed by SuperSonic Imagine. Delay generator: low jitter digital delay generator, Stanford Research Systems. AWG: arbitrary waveform generator, Tektronics AWG7010. Generate pre-loaded customized waveform with a 1.2 GHz reading precision.

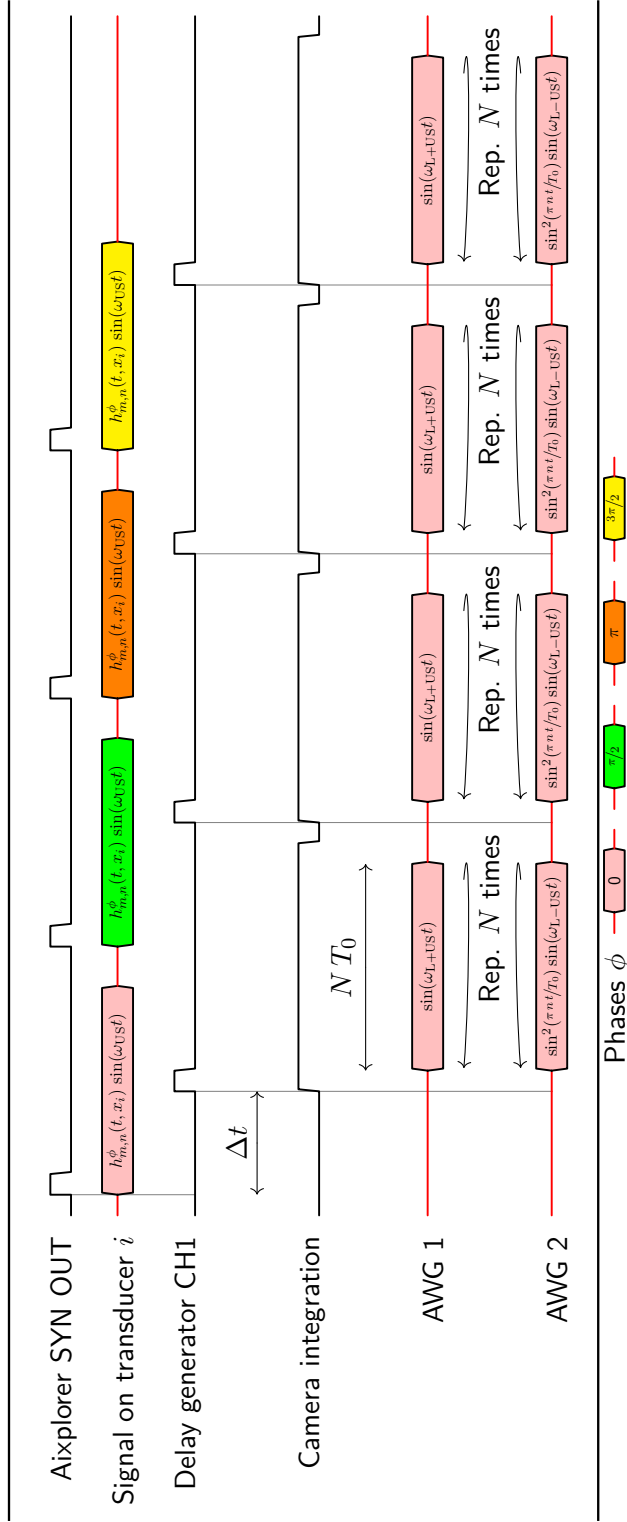


Figure C.3 – Chronogram of the devices involved in the Fourier transform imaging setup for a given example of structuration of indices m, n . $\omega_L + \omega_{US} = \omega_L + \omega_{US}$ and $\omega_L - \omega_{US} = \omega_L - \omega_{US}$. Vertical drawn lines highlight the synchronicity driven architecture presented on Figure C.2. The camera integration period is set larger than the temporal width of the modulation on AWG1 and AWG2. This is due to the uncertainty on actual integration time applied by the camera. Indeed, this integration time could be different from the set value. On the contrary, the arbitrary waveform generator has a GHz reading precision. This ensure the same temporal length for all the modulations. The signal displayed in boxes refer to the waveform described in subsection 2.4.2. The delay Δt applied by the trig generator is chosen to correspond to the time of flight of the ultrasonic wave to the region of interest.

C.2 AOTM measure experimental setup

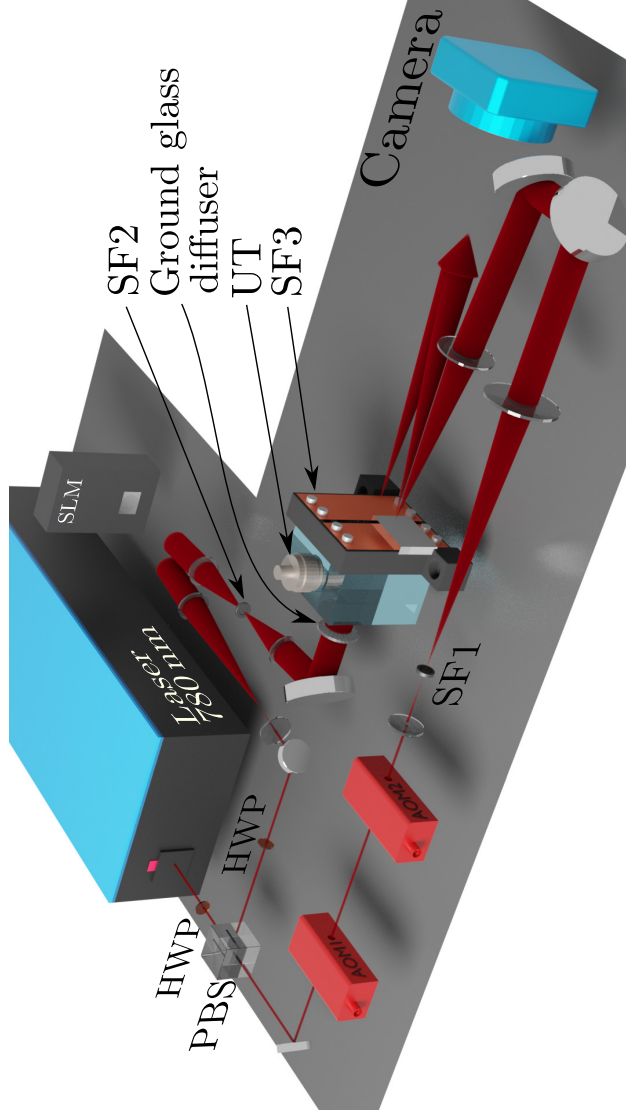


Figure C.4 – Schematic of the optical setup used to measure the AOTM. The continuous laser at 780 nm is split in two beams through a polarized beam splitter, the power ratio between the two arms is adjusted with a half-wave plate (HWP). (SF#) Spatial filters: (SF1) is a pin hole with a 200 μm diameter, (SF2) is a width-adjustable slit and (SF3) is the slot used to limit the numerical aperture of the speckle for off-axis holography, (UT) ultrasonic transducer.

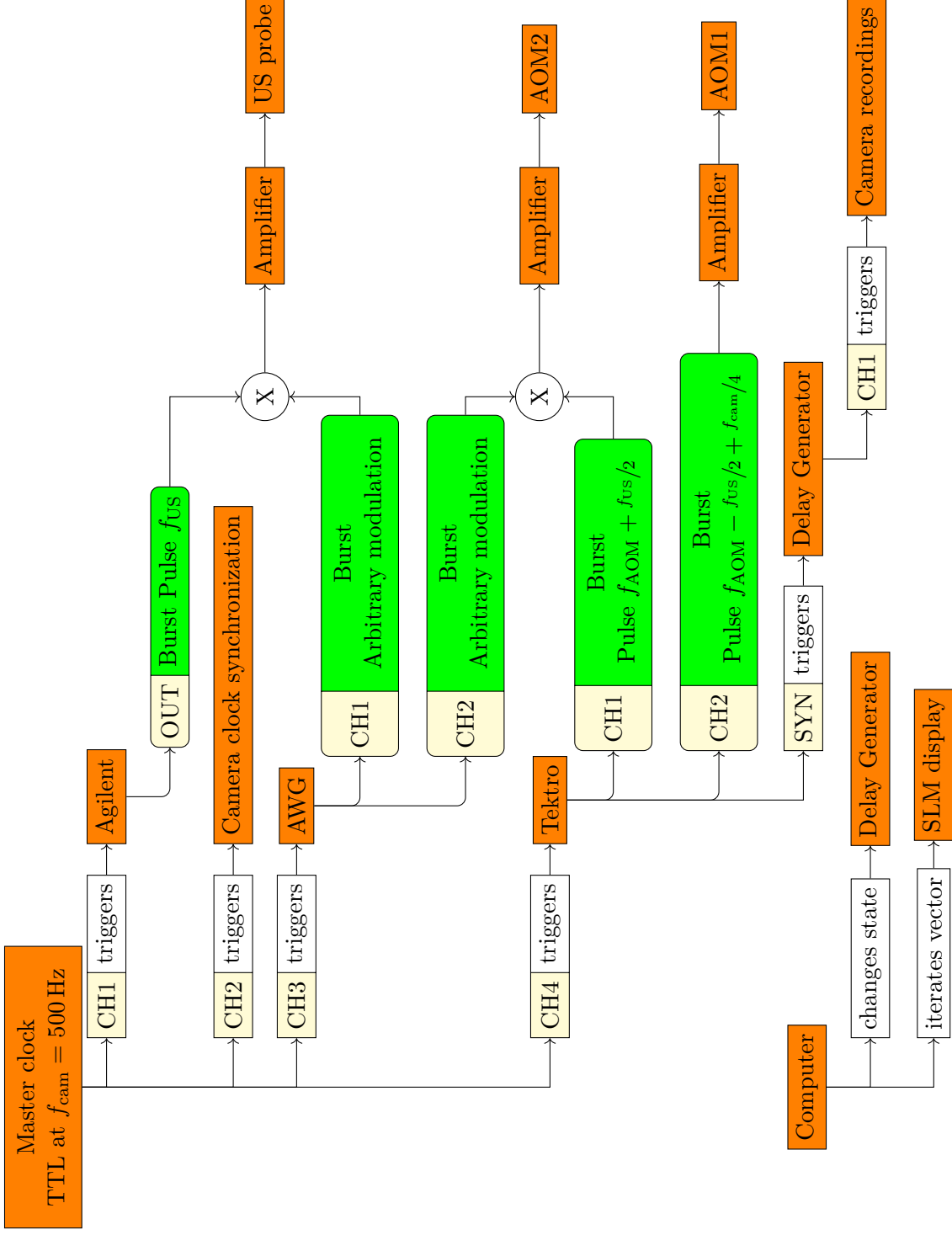


Figure C.5 – Electric connection of the AOTM measurement setup, corresponding to a four-phase temporal filtering method. Master clock: Digital delay generator, Stanford Research Systems, emits clock signals at 500 Hz (frame rate of the camera) to trigger the other generator with adapted delays. US probe: OLYMPUS PARAMETRICS -NDT- A307S of central frequency 5 MHz and a fixed focal distance of 50 mm. Agilent: sinusoidal wave generator in burst mode. Tektro: Tektronics' waveform generator in burst mode. Delay generator: digital delay generator used as an on/off remotely switchable relay for triggering signal to the camera. X denotes the signal multiplier.

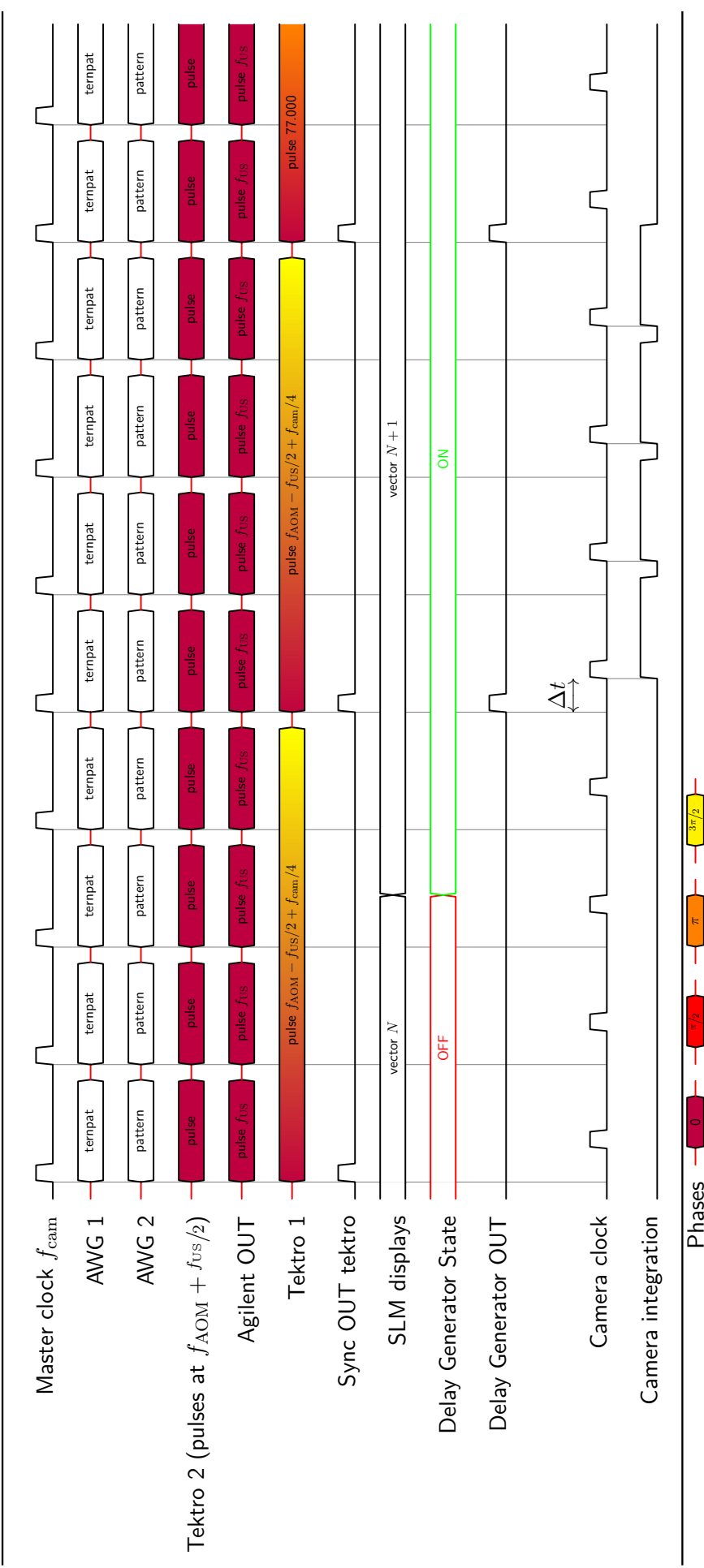


Figure C.6 – Chronogram of the devices implied in the AOTM measurement setup. Vertical drawn lines highlight the synchronicity driven by the connection architecture presented on Figure C.5. The camera clock cycles are shifted with a temporal delay Δt , this delay corresponds to the time of flight of the US across the sample. The fast camera is in ‘random 4’ recording mode. In this mode the camera waits for a triggering event and then records 4 frames corresponding to the four successive cycles of its internal clock. As its internal clock is locked with the pulse generator which also triggers the waveform generator, the frames recorded by the camera are all in phase. All the waveform generators have their internal clock synchronized at 120 MHz. The Arbitrary Waveform Generator (AWG) generates on its two output channels, AWG 1 and AWG 2, the same waveform pattern. The waveform, for instance a pattern of successive random phase shifts is temporally rotated ‘termpat’ on AWG 1. The angle of this rotation is tuned to match the delay imposed by the time of flight of the ultrasounds from the probe to the region of interest within the sample.

ANNEXE D

Résumé en français

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Introduction

L'IMAGERIE est un outil clef du diagnostic médical, donnant accès à des éléments essentiels à la compréhension des phénomènes physiologiques et métaboliques. La dépistage précoce étant d'une importance cruciale pour le succès des traitements de plusieurs maladies, la recherche actuelle en imagerie s'appuie à repousser les limites de résolution des méthodes existantes. Ces progrès vont de paire avec l'amélioration de la sensibilité offrant une information plus claire sur laquelle le médecin peut fonder son diagnostic. La portabilité de l'imagerie médicale contribue aussi à la détection précoce. Par exemple, le développement de systèmes d'échographie compacts a permis leur implantation dans les cabinets médicaux ou les véhicules d'urgence, les rapprochant ainsi des patients. De plus, l'évolution vers des méthodes d'exploration moins invasives diminue les répercussions sur le patient et la durée des interventions. La diversité des techniques d'imagerie apporte des informations complémentaires comme autant de points de vue différents sur une pathologie.

Dans ce contexte, les méthodes d'imagerie optique proposent de nombreux avantages. Elles sont pour la plupart non-invasives, les endoscopes utilisent des fibres optiques pour observer à l'intérieur du corps à la surface des organes, et avec une grande résolution, sub-micrométrique pour les microscopes, qui permettent de suivre l'activité interne des cellules. Une grande variété de techniques optiques peuvent être mises à profit pour observer les phénomènes biologiques et ainsi comprendre des fonctions métaboliques. En effet, les marqueurs biologiques, appelés chromophores, tels que l'hémoglobine (protéine vectrice de l'oxygène), absorbent la lumière à différentes longueurs d'onde. Nous pouvons donc lier les fonctions métaboliques à la mesure de l'absorption dans les tissus. Ainsi, l'optique permet une imagerie vasculaire et par conséquent fonctionnelle des organes. La propagation de la lumière au travers des milieux biologiques est aussi caractérisée par la diffusion multiple. Au-delà d'un millimètre de profondeur dans les tissus, la lumière a perdu la mémoire de sa direction et de sa polarisation initiales. Ce phénomène s'impose comme limite aux techniques d'imagerie optique standards telles que la microscopie ou la tomographie de cohérence optique (OCT) dans la plupart des tissus du corps humains.

L'imagerie multi-onde apporte une solution à la limite d'observation imposée par la diffusion. Fondées sur le couplage entre lumière et ultrasons, la tomographie photo-acoustique (PAT) et l'imagerie acousto-optique (AOI) bénéficient des avantages combinés de l'échographie et de la mesure d'un contraste optique. En effet, les ultrasons ont une propagation balistique dans les tissus, ce qui permet une émission et réception à distance sans perte de la résolution acoustique, et le contraste optique permet de remonter à des informations biologiques et métaboliques. L'AOI utilise des ultrasons pour guider la lumière au-delà d'un millimètre de profondeur. Cette méthode consiste à focaliser une onde acoustique dans l'échantillon éclairé par une source laser. Dans la zone d'interaction des ultrasons et du faisceau lumineux, une partie des photons sont spectralement décalés de la fréquence des ultrasons. Par conséquent, tout ce passe comme si l'on disposait de sources virtuelles situées à des positions connues à l'intérieur du milieu. Comme l'amplitude de ces sources virtuelles est proportionnelle à l'absorption locale, la détection des photons

marqués donne accès au contraste optique dans la zone d'interaction entre la lumière et les ultrasons. En balayant la tache focale des ultrasons, on peut ainsi construire une carte des propriétés optiques de l'échantillon, telle que l'absorption ou la diffusion.

La principale difficulté de l'AOI concerne la détection. En effet, la contribution des photons est spectralement très proche de la lumière non marquée, ce qui rend impossible leur résolution spectrale par des spectromètres conventionnels. Plusieurs techniques de filtrages de la contribution des photons marqués ont été développées. Dans ce travail, nous utilisons l'holographie numérique. La lumière diffusée est collectée sur le capteur d'une caméra, ce qui rend cette technique relativement légère à mettre en œuvre. La contribution de la lumière non-marquée est séparée en utilisant un oscillateur local à la fréquence des photons marqués. En tant que méthode interférométrique, cette méthode repose sur une illumination cohérente, elle est donc sensible aux décorrélations, spatiale et temporelle, de la lumière qui interviennent dans les milieux biologiques. En effet, à cause du mouvement stochastique des structures à différentes échelles, la figure d'interférence captée par la caméra est renouvelée après un temps caractéristique d'une milliseconde. Dans un but d'imagerie *in vivo* la détection de la lumière marquée doit donc être effectuée en moins d'une milliseconde. Pour cela, une méthode d'excitation acoustique par des ondes planes structurées a été proposée produisant suffisamment de signal marqué pour être robuste à la décorrélation temporelle.

De plus, l'holographie numérique donne accès à la phase du front d'onde optique des photons marqués. Ensuite, en utilisant un modulateur spatial de lumière (SLM), il est possible de contrôler le front d'onde incident au milieu. La diffusion au travers de l'échantillon diffusant peut être assimilé à un processus linéaire liant des composantes optiques d'entrée et de sortie, le marquage acousto-optique jouant le rôle de contrainte spatiale sur la propagation. Pour cela, on applique la théorie des matrices de transmission pour identifier les canaux optiques les plus transmissifs. Cette thèse donne des approches matricielles de l'AOI avec un premier axe sur l'excitation acoustique suivi d'un second sur les canaux de transmission acousto-optique à travers le système optique.

Ce travail est organisé en quatre parties. La première partie présente la propagation de la lumière dans les milieux biologiques. Nous y exposons les différentes méthodes d'imagerie, entièrement optiques ou hybrides ; la tomographie optique diffuse et la PAT. Après avoir présenté les mécanismes de l'effet acousto-optique, nous démontrons comment la détection par holographie sur caméra permet de sélectionner la lumière marquée. La seconde partie se concentre sur l'excitation acoustique. Nous développons différentes modulations temporelles utilisables avec des ultrasons focalisés. Puis nous nous intéressons à des réseaux d'émetteurs et aux ultrasons spatialement structuré, pour aller vers une approche matricielle de l'excitation. La troisième partie présente les techniques de façonnages du front d'onde optique et introduit les matrices de transmission. Nous expliquons en quoi les matrices de transmission constituent un outil pour focaliser la lumière dans un milieu diffusant par retournement temporel. Nous finissons par décrire les singularités de la matrice de transmission acousto-optique (AOTM). La partie finale présente la mesure de l'AOTM puis explique comment les vecteurs singuliers permettent de focaliser plus finement que la limite de diffraction acoustique.

D.1 Imagerie optique dans les milieux diffusants

Dans cette première section, nous nous intéressons à décrire la propagation de la lumière dans les milieux biologiques. Nous présentons ensuite des méthodes optiques et hybrides d'imagerie dans ces milieux. Enfin, nous verrons comment il est possible de sélectionner la contribution des photons marqués de la lumière diffusée en sortie du milieu.

D.1.1 Optique dans les milieux biologiques

La première contrainte à laquelle l'imagerie optique dans les milieux biologiques se confronte est l'absorption. Suivant la loi de Beer-Lambert, l'intensité lumineuse décroît exponentiellement avec la profondeur. Cette décroissance varie selon les éléments absorbants constitutifs du milieu et est caractérisée par le coefficient d'absorption μ_a . Ce coefficient est spécifique de chaque chromophore, i.e. éléments constitutifs des milieux biologiques optiquement actifs. Les principaux chromophores sont l'hémoglobine et ses formes oxydées, l'eau, le collagène, etc. L'activité des chromophores dépend de la longueur d'onde de la lumière. Aussi existe-t-il une plage du spectre lumineux, appelée fenêtre optique thérapeutique de 600 nm à 1300 nm, pour laquelle l'absorption globale du milieu est minimale et donc où la lumière pénètre le plus en profondeur (voir [figure 1.1](#) page 21).

Le second phénomène affectant la propagation de la lumière dans les milieux biologiques est la diffusion. Ce phénomène est dû aux hétérogénéités d'indice optique dans le milieu, qui ont pour effet de dévier les différents chemins de la lumière de façon non prévisible. Ceci a pour effet de rendre inaccessible la localisation de l'information optique. La diffusion est caractérisée par le libre parcours moyen de diffusion qui correspond à la distance moyenne entre deux événements de diffusion dans le milieu l_s qui vaut typiquement 100 μm dans les milieux biologiques.

Cette diffusion se fait statistiquement dans une direction privilégiée, cette anisotropie est quantifiée par le facteur g . Le libre parcours moyen de transport l^* tient compte de la propagation de la lumière en marche aléatoire. Cette quantité correspond à la distance au-delà de laquelle la lumière a perdu la mémoire de sa direction et de sa polarisation initiales. Elle vaut typiquement 1 mm dans les tissus biologiques. Les propriétés optiques des tissus biologiques ont été mesurées par plusieurs études, le [tableau 1.1](#) page 24 fait le bilan de ces mesures.

Selon l'épaisseur W du milieu traversé par la lumière, différents régimes de propagation existent :

- pour $W \ll l^*$, la lumière interagit très peu avec les diffuseurs. La propagation est dominée par le régime balistique, domaine de validité des méthodes optiques standard telles que la microscopie ou la tomographie de cohérence optique (OCT).
- pour $W \sim l^*$, la lumière suit un régime de diffusion simple. Le faisceau s'étend selon un cône, la lumière suit des chemins sinueux à travers le milieu.
- pour $W \gg l^*$, on entre dans le régime de diffusion multiple. Tous les photons subissent de nombreux événements de diffusion. Dans les milieux biologiques, ce

régime intervient au-delà d'un millimètre de profondeur.

D.1.2 La figure de *speckle*

En sortie d'un milieu diffusant illuminé par un faisceau monochromatique, le faisceau optique est très élargi. Du fait des multiples chemins optiques empruntés par la lumière, la phase de ce faisceau est spatialement hétérogène. La figure d'auto-interférence produite par cette onde est appelée *speckle* (ou tavelure), la [figure 1.3](#) (page 25) en donne une représentation. Cette figure montre une succession de zones brillantes, appelées grains de *speckle*, dont la taille caractéristique est donnée par la longueur d'onde du faisceau et l'ouverture numérique de la tache de diffusion dans le plan d'observation (voir [équation 1.4](#) page 25).

D.1.3 Techniques d'imagerie optiques

Plusieurs techniques d'imagerie optiques des milieux biologiques existent. Dans cette section, nous nous concentrons sur les techniques applicables en régime de diffusion multiple.

Tomographie optique diffuse Cette méthode d'imagerie repose sur une émission et détection de la lumière par un réseau de capteurs. L'ensemble des détecteurs enregistre les flux correspondants aux émissions individuelles et successives de chacun des émetteurs. La carte d'intensité est par la suite reconstruite par un algorithme. Cette méthode permet une imagerie résolue au millimètre sur une profondeur limite de 6 cm ([Dalla Mora et al., 2015](#)).

Tomographie photo-acoustique Cette méthode multi-onde est souvent confondue avec l'imagerie acousto-optique. Elle consiste à éclairer le milieu avec un laser pulsé (*Q-switch*). L'absorption du faisceau par les chromophores est dissipé sous forme de rayonnement thermique. L'élévation de la température provoque une dilatation locale du milieu, créant ainsi une onde mécanique dans la plage du mégahertz (voir [figure 1.4](#) page 28). Le profil temporel de cette onde acoustique est enregistrée par une sonde d'échographie à la surface du milieu. L'analyse des signaux acoustiques permet ainsi de remonter à la distribution de l'énergie optique absorbée par les tissus ([Jayet, 2015](#)).

D.1.4 Imagerie acousto-optique

L'imagerie acousto-optique se fonde sur la détection de photons marqués spectralement par une onde acoustique. Ce marquage est le résultat de l'effet acousto-optique qui intervient lorsque des ultrasons croisent le chemin d'un faisceau lumineux. En effet l'onde mécanique en propagation agit comme une succession de compression et relaxation locale du milieu à la fréquence des ultrasons. Cet effet mécanique modifie légèrement l'indice optique du milieu, l'onde acoustique va donc créer un réseau d'indice dynamique sur son passage. En conséquence, une partie des photons qui traversent le passage des

ultrasons va être décalée spectralement de la fréquence des ultrasons (voir le schéma de la [figure 1.6](#) page 30). L'intensité des photons marqués est proportionnelle à l'intensité lumineuse locale intégrée sur le volume occupé par les ultrasons. La détection sélective des photons marqués permet donc de sonder le contraste optique à l'intérieur de la tache focale acoustique avec une résolution théoriquement définie par la limite de diffraction acoustique. Une carte de ce contraste est reconstruite en déplaçant la tache focale acoustique.

Creusement de trous spectraux Du fait de la différence de plusieurs ordres de grandeur entre le décalage spectral acousto-optique et la fréquence centrale du faisceau sonde, la détection nécessite un filtrage spectral très étroit. De tels filtres peuvent être créés par creusement de trous spectraux, ils nécessitent d'être refroidis à des températures cryogéniques. Ils consistent en des cristaux dopés aux terres rares dont la raie d'absorption couvre à la fois le spectre des lumières marquée et non-marquée par les ultrasons. Un faisceau pompe à la fréquence des photons marqués est utilisé pour saturer une raie homogène du spectre d'absorption du cristal (voir schéma de la [figure 1.7](#) page 31). La raie homogène des ions de terres rares utilisés pour ces filtres étant fine de l'ordre du kilohertz à des températures cryogéniques, par exemple 3 kHz à 1.5 K dans le Tm:YAG ([Venet, 2019](#)), le creusement de trous spectraux permet de créer une fenêtre de transparence suffisamment étroite pour ne laisser passer seulement la lumière marquée par les ultrasons. La détection par creusement de trous spectraux est fondée sur une mesure de flux lumineux, ce qui la rend robuste à la décorrélation temporelle du milieu observé.

Holographie numérique Dans l'ensemble des travaux développés dans cette thèse nous avons utilisé l'holographie hétérodyne hors-axe comme méthode de détection. Il s'agit d'une méthode d'interférométrie. Le *speckle* issu du faisceau sonde est recueilli en champ lointain sur une caméra. Le capteur de la caméra est aussi éclairé par un faisceau référence, à la fréquence des photons marqués, qui joue le rôle d'oscillateur local. La contribution des photons marqués est filtrée par une méthode temporelle, en détection hétérodyne, ou spatiale, franges d'interférence dans la bande passante de la caméra. Dans la [sous-section 1.6.3](#) (page 37) nous développons le filtrage spatial, méthode sur laquelle nous nous sommes appuyés.

Dans cette section, nous avons vu pourquoi les techniques d'imagerie optique sont cruciales pour le diagnostic et la détection précoce de certaines maladies. Après avoir développé la raison pour laquelle la diffusion multiple impose une limite aux techniques d'imagerie optique standard, nous avons montré que le modèle de diffusion complexe qui émerge en éclairant un tel milieu contient toujours des informations sur la source de lumière et son histoire. L'effet acousto-optique permet de marquer la lumière passant dans une région spécifique. L'amplitude de la lumière marquée est directement liée au contraste local dû à l'absorption et à la diffusion. Enfin, nous avons présenté l'holographie numérique qui permet spécifiquement la détection du signal acousto-optique. Tous ces éléments réunis constituent l'imagerie acousto-optique en détection sur caméra.

D.2 Mise en forme de l'excitation acoustique

Dans cette partie nous développons les modulations acoustiques utilisées en imagerie acousto-optique. Comme nous l'avons vu dans la section précédente, la détection des photons marqués permet de remonter au contraste optique dans la tache focale des ultrasons.

D.2.1 Ultrasons focalisés

En pratique les ultrasons sont émis par des transducteurs uniques sphériques. Ces transducteurs produisent une tache focale de forme allongée. Cette géométrie fournit une localisation dans le plan transverse à la propagation des ultrasons (voir [figure 2.2](#) page 50). Pour avoir un sectionnement le long de l'axe de propagation, une solution simple est d'utiliser des impulsions combinées avec des temps d'intégration courts sur la caméra. En prenant des images successives, on peut ainsi suivre la progression de l'impulsion à travers un échantillon limpide tel que présenté sur la [figure 2.3](#) page 51. Néanmoins, la résolution étant proportionnelle à la longueur de l'impulsion et au temps d'intégration de la caméra, ces durées doivent rester de l'ordre le microseconde pour résoudre au millimètre, ce qui réduit par là-même la quantité de photons marqués collectés. Dans le cas de l'imagerie d'un milieu fortement diffusant pour laquelle la quantité de signal est un enjeu limitant, cette méthode n'est pas assez efficace. C'est pourquoi des méthodes de modulations sur des longues impulsions d'ultrasons, de 50 μs à quelques millisecondes, ont été implémentées.

Une de ces méthodes, présentée pour la première fois par [Lesaffre et al. \(2009\)](#), met à profit les propriétés d'auto-corrélation des signaux aléatoires. Une longue impulsion ultrasonore est modulée par une suite aléatoire de 1 et -1 de durée égale, modulation appelée sauts aléatoires de phase. Lorsque cette modulation (voir [figure 2.6](#) page 55) est appliquée à la fois sur les ultrasons et sur le faisceau référence en configuration d'holographie hétérodyne hors-axe, l'intégration de la caméra joue le rôle d'une auto-corrélation du motif aléatoire. L'étalement du pic central de cette auto-corrélation est égal à la largeur des bascules de phase employées, permettant ainsi un sectionnement le long de l'axe de propagation des ultrasons tout en intégrant le signal sur une longue période. Cette modulation a été mise en œuvre expérimentalement pour la première fois par [Lesaffre \(2011\)](#) en détection photoréfractive, puis par [Benoît à la Guillaume et al. \(2012\)](#) en holographie hétérodyne hors-axe.

La modulation par des bascules aléatoires de phase nous a permis d'imager des inclusions dans un gel fortement diffusant (intralipid-agar), $\mu'_s = 10 \text{ cm}^{-1}$ de 2 cm d'épaisseur (voir [figure 2.14](#) page 64) et d'imager la diffusion du faisceau sonde au travers de l'échantillon (voir [figure 2.15](#) page 64). Néanmoins, cette méthode requiert l'utilisation de signal long proche de la milliseconde, ce qui la rend incompatible avec l'imagerie *in vivo*. De plus, chaque mesure doit être moyennée sur plusieurs centaines d'acquisitions pour améliorer le rapport signal à bruit, ce qui donne une durée d'imagerie de l'ordre de plusieurs minutes ou dizaines de minutes.

D.2.2 Ondes planes ultrasonores

Pour pallier à la faiblesse du signal produit par les ultrasons focalisés, [Laudereau et al. \(2016\)](#) ont proposé d'utiliser des ondes planes ultrasonores. En effet en remplaçant le transducteur mono-élément par une barrette d'émetteurs indépendants, on élargit la zone de marquage acousto-optique, ajoutant ainsi deux ordres de grandeurs à son volume (voir l'application numérique de l'équation 2.31 page 66). Le rapport signal à bruit étant meilleur, on peut alors s'affranchir de moyenner le signal mesuré sur des centaines d'acquisition et ainsi accélérer l'imagerie d'un facteur 50. La reconstruction de l'image est différente de celle des ultrasons focalisés pour laquelle la base de mesure correspond au pixels de l'image finale. [Laudereau et al. \(2016\)](#) utilise des ondes planes inclinées de $\pm 20^\circ$ en imposant une loi de retard sur les émissions des transducteurs de la barrette ultrasonore. Cette limite angulaire induit une anisotropie de résolution entre l'axe de propagation des ultrasons et la direction transverse. Par pallier à cela [Laudereau et al. \(2016\)](#) réalise deux séries de mesures en inclinant la barrette à 90° (voir figure 2.16 page 67).

Cette technique d'ondes planes a été améliorée par [Bocoum et al. \(2019\)](#). Des modulations d'amplitude à différentes périodes le long de l'axe de la barrette ultrasonore sont ajoutées pour résoudre les détails dans la direction transverse (voir figure 2.17 page 68). On peut alors imager, sans déplacer la sonde ultrasonore, avec une résolution imposée par la limite de diffraction acoustique.

D.2.3 Imagerie par transformée de Fourier

Lors de cette thèse, nous avons utilisé l'imagerie par transformée de Fourier. Cette méthode consiste à remplacer les ondes planes pulsées de la méthode précédente par une série périodique d'impulsions. En jouant à la fois sur les périodes, spatiale grâce à la modulation appliquée le long de l'axe de la barrette, et temporelle avec la série d'impulsions, on peut ainsi créer une base de motifs d'ultrasons (voir le schéma de la figure 2.19 page 69). La mesure du signal acousto-optique produit par chaque motif permet d'attribuer un coefficient à la composante de Fourier de l'image correspondant aux périodes spatiales et temporelles du motif de modulation (voir figure 2.20 page 70). Cette nouvelle méthode est présentée combinée à une détection par holographie photoréfractive dans ([Bocoum et al., 2020](#)). En détection par holographie numérique ([Dutheil et al., 2021](#)), le temps d'intégration de la caméra est un multiple de la durée d'un motif, augmentant ainsi la quantité de signal. On a ainsi pu réaliser une mesure de chacune des composantes de Fourier en une seule mesure. Les images présentées figure 2.27 page 78 et figure 2.29 page 79 ont été réalisées dans des milieux diffusant de coefficients de diffusion réduits respectifs de 1 cm^{-1} et 10 cm^{-1} en 1.5 s.

Ces intégrations spatiales et temporelles plus importantes permettent de collecter suffisamment de signal pour réaliser une acquisition complète sans avoir à moyenner les mesures sur un grand nombre d'acquisition. Dans cette condition, l'imagerie est robuste à la décorrélation rapide des tissus vivants. De plus, l'utilisation d'une onde plane US et d'une impulsion de longue durée limite la puissance de crête délivrée dans l'échantillon,

permettant ainsi de rester en-deçà du seuil des normes biomédicales. Enfin, le temps d'acquisition global est de 1.5 s, ce qui rend cette méthode adaptée au suivi *in vivo*.

Dans cette section, nous avons présenté les différentes stratégies de modulation qui ont été développées pour l'imagerie acousto-optique. Nous avons vu que l'utilisation d'une sonde d'échographie fournit une large excitation acoustique permettant d'imager un plan dans un temps d'acquisition global compatible avec les contraintes de l'imagerie *in vivo*. La FT-AOI est prometteuse pour de futures investigations dans les tissus vivants. De plus, des améliorations sont prévisibles dans la reconstruction des images. En effet, nous avons vu lors de simulations que certains motifs ultrasonores sondent plusieurs composantes de Fourier du plan imagé en même temps. Pour l'instant, ceci est une limitation de la diversité des motifs utilisables en FT-AOI. Néanmoins, nous pouvons profiter de ces effets de diffraction en mesurant les fonctions de marquage pour chaque motif dans un environnement connu, par exemple l'observation balistique dans une cuve d'eau. Nous pouvons ensuite adapter la reconstruction en tenant compte de tout le spectre de Fourier bidimensionnel du motif de marquage, y compris les effets des bords du motif et de la diffraction acoustique. Pour ce faire, nous simulons la fonction de marquage dans le plan ultrasonique pour chaque motif structuré. Cela nous permettrait de construire une matrice des composantes spatiales-spectrales de chaque motif structurant dans le domaine spatial de Fourier. L'inversion de la matrice permettrait d'améliorer le remplissage des composantes de Fourier du plan imagé. Ensuite, en utilisant les opérations d'analyse matricielle que nous verrons dans la section suivante, nous pourrions concevoir un motif de base basé sur la composante principale d'une telle matrice de marquage acousto-optique.

D.3 Focaliser la lumière dans des milieux diffusants

Comme nous l'avons vu dans les sections précédentes, l'imagerie acousto-optique a été considérablement améliorée par l'utilisation de réseaux de transducteurs. La multiplication des émetteurs offre ainsi un nouveau degré de liberté et de nouvelles possibilités d'échantillonnage du contraste optique au sein du milieu. Du côté optique, le développement de dispositifs numériques pour moduler le front d'onde optique permet de sonder les canaux de transmission et de réflexion d'un échantillon diffusant. Dans cette section, nous allons montrer comment les techniques de modulation du front d'onde optique peuvent être utilisées pour mesurer la matrice de transmission optique des milieux diffusants.

D.3.1 Contrôle du front d'onde optique

Les dispositifs optiques de correction de phase ont d'abord été développés en astronomie pour compenser la distorsion des images d'observation due à l'atmosphère. Cette technique est mise en œuvre sur la plupart des télescopes et communément appelé optique adaptative. Cette technique a ensuite été transposée pour des applications en microscopie, en particulier pour l'imagerie de l'œil afin de compenser les aberrations oculaires. Néanmoins, l'optique adaptative atteint ses limites pour le régime des fortes aberrations car le nombre de modes à contrôler dépasse les capacités des miroirs déformables. Au cours des quinze dernières années, les techniques de mise en forme du front d'onde sont apparues comme un outil de choix pour contrôler la propagation de la lumière à travers des milieux diffusants, en particulier pour les applications d'imagerie (Popoff et al., 2010b; Xu et al., 2011; Mosk et al., 2012; Judkewitz et al., 2013; Katz et al., 2014; de Aguiar et al., 2016). La mise en forme du front d'onde consiste à moduler spatialement l'amplitude et/ou la phase du faisceau incident sur un milieu de propagation donné. Trois modulateurs spatiaux de lumière sont couramment utilisés :

- les modulateurs spatiaux de lumière à cristaux liquides sur silicium (LCOS-SLM). Cette technologie est basée sur les propriétés de biréfringence des cristaux liquides (Savage, 2009). Les cellules à cristaux liquides sont constituées d'une couche de cristaux liquides incluse entre deux électrodes commandant l'orientation des molécules. Ainsi l'indice apparent de la cellule est pilotée par la tension appliquée sur les électrodes. Les LCOS-SLM fournissent ainsi une modulation de phase de 0 à 2π à une longueur d'onde donnée avec environ un million de degrés de liberté. Leur temps de réponse des cellules à cristaux liquides ($\sim 20 \mu\text{s}$) limite sa vitesse à quelques dizaines de hertz. Dans cette thèse, nous utilisons un LCOS-SLM modulant la phase en réflexion.
- Les MEMS-SLM sont similaires aux miroirs utilisés en optique adaptative, comme le montre la figure 3.2 page 83. Ils consistent en un réseau de miroirs plats de $10 \mu\text{m}$ à $100 \mu\text{m}$ montés sur des actionneurs linéaires individuels contrôlés par des forces électrostatiques. Par conséquent, l'élévation de chaque miroir induit une modulation de 0 à au moins 2π pour une longueur d'onde donnée. La faible temps de réponse des actionneurs, $10 \mu\text{s}$ à $100 \mu\text{s}$, permet à cette technologie d'atteindre

un taux de rafraîchissement 10 kHz à 100 kHz.

- Les DMD sont composés de miroirs inclinables d'un pas de 10 μm à 100 μm , comme le montre la [figure 3.3](#) page 84. Chacun d'entre eux peut basculer de manière indépendante et contrôlable entre deux angles lorsqu'une tension est appliquée. Ils fournissent une modulation d'amplitude binaire dans une direction de l'espace. Cette technologie peut atteindre un taux de rafraîchissement de quelques dizaines de kilohertz.

Le [tableau D.1](#), ci-dessous, résume les principales caractéristiques de ces SLM couramment utilisés. Dans la section suivante, nous présentons la première expérience dé-

Technologie	LCOS-SLM	MEMS	DMD
Nombre de pixels	10^6	10^3	10^5
Taux de rafraîchissement (Hz)	$10^{1\cdots 2}$	10^5	$10^{4\cdots 5}$
Modulation	En phase $0 \cdots 2\pi$	En phase $0 \cdots 2\pi$	Binaire en amplitude on/off
Prix (€)	10^4	10^5	10^3

Table D.1 – Tableau comparatif des technologies de contrôle du front d'onde optique.

montrant la focalisation de la lumière à travers un milieu diffusant.

D.3.2 Focaliser la lumière derrière une couche diffusante

Dans une expérience fondatrice ([Vellekoop and Mosk, 2007](#)), [Vellekoop et al. \(2010\)](#) ont réussi à focaliser la lumière à travers un milieu diffusant en utilisant la mise en forme du front d'onde. Cette expérience consiste à itérer sur les valeurs de modulation de phase d'un front d'onde optique d'entrée afin de compenser l'effet de la diffusion pour concentrer la lumière en un point précis (voir [figure 3.4](#) page 85). Dans cette expérience, le SLM est divisé en N segments individuels. En partant d'un masque de phase plat, la valeur de la phase de chaque segment est décalée de 0 à 2π . La phase correspondant à l'intensité la plus forte dans la zone cible est conservée (voir [figure 3.5](#) page 86). L'optimisation passe ensuite au segment suivant. Le facteur d'amélioration global η de cette méthode est défini comme le rapport entre l'intensité optimisée et l'intensité moyenne initiale et évolue avec le nombre de degrés de liberté N , c'est-à-dire le nombre de segments sur le SLM ([Vellekoop et al., 2010](#)) :

$$\eta_{\text{eff}} = \frac{\pi}{4}(N - 1) + 1. \quad (\text{D.1})$$

Une telle technique basée sur une optimisation par une boucle de rétro-contrôle sur le champ incident s'est avérée efficace pour focaliser la lumière à travers un milieu diffusant. Cependant, elle nécessite d'avoir accès aux deux côtés du milieu. La principale difficulté de la focalisation dans un milieu diffusant est que nous ne pouvons pas accéder directement à la zone cible. Pour cette raison, les méthodes qui ont été développées au

cours de la dernière décennie impliquent ce que l'on appelle un composant *guide-star* plongé dans le milieu, interagissant spécifiquement avec l'illumination. Pour être utilisé, le résultat de cette interaction doit être détectable de l'extérieur du milieu afin de mettre en œuvre un contrôle par rétroaction.

D.3.3 Focalisation de la lumière dans un milieu diffusant

Le nom *guide-star* provient de l'optique adaptative en astronomie, lorsque [Fugate et al. \(1991\)](#) utilisait un laser pour créer un point de repère étroit dans l'atmosphère. Le concept de cette technique est d'utiliser un objet cible aux propriétés connues (forme, phase, absorption) noyé dans le milieu d'étude. L'observation de l'interaction entre cette cible et le front d'onde optique envoyé révèle des informations sur les propriétés de propagation entre le plan d'entrée et la cible.

Acousto-optique comme méthode *guide-star* Dans ce contexte, l'acousto-optique offre une interaction locale entre la lumière et le son à l'intérieur de milieux diffusants. La prédominance du régime balistique pour les ondes ultrasonores dans les hydrogels et les milieux biologiques nous permet de contrôler la forme de cette interaction ([Lerosey and Fink, 2013](#); [Ma et al., 2015](#)). De plus, elle n'implique pas l'introduction d'une sonde ou de marqueurs à l'intérieur du milieu ce qui la rend non invasive et facilement réalisable. L'observation sélective du champ optique marqué offerte par l'holographie numérique permet d'accéder à la quantité de lumière traversant le foyer ultrasonore.

Conjugaison de phase optique et retournement temporel Dans les années 90, Mathias Fink a initié les techniques de retournement temporel ([Fink, 1992, 1997](#)). Développée d'abord en acoustique ([Prada et al., 1996](#)) au laboratoire 'ondes et acoustique', la technique a été transposée à d'autres domaines de la physique des ondes; micro-ondes ([Lerosey et al., 2007](#); [Fink et al., 2013](#)), optique ([Popoff et al., 2011](#)) et a offert de multiples applications notamment pour l'imagerie et les télécommunications. La technique est basée sur la réversibilité de l'équation d'onde, ce qui signifie que la propagation d'une onde est inchangée en inversant le cours du temps. Elle s'inspire également du théorème de Helmholtz-Kirchhoff qui stipule que la connaissance simultanée d'un champ et de sa dérivée en tous points d'une surface fermée, permet de retrouver le champ en tout point du volume délimité. Imaginons une source ponctuelle émettant une onde temporellement courte, grâce au théorème de Helmholtz-Kirchhoff, si nous installons un réseau continu de capteurs autour de la source, enregistrant le passage de l'onde, inversant temporellement le signal et le restituant, la somme de toutes les ondes réémises se concentrera à la position de la source, à la fois spatialement et temporellement. Expérimentalement, une telle expérience est difficile à mettre en œuvre. En effet, pour avoir accès à une surface fermée autour des sources avec des transducteurs espacés de la moitié de la longueur d'onde, le nombre total de capteurs serait énorme. De plus, la distribution spatiale des capteurs permet un échantillonnage spatio-temporel discret. En pratique, nous n'avons accès au champ que sur une surface limitée, ce qui limite

l'ouverture numérique effective du système. En optique monochromatique, l'opération de retournement temporel correspond à une conjugaison de la phase du champ optique. Elle permet une focalisation spatiale de la lumière.

Approche matricielle du retournement temporel Pour un milieu linéaire, la relation entre le champ optique dans un plan donné et celui dans un plan de sortie peut être décrite par une matrice. Une telle matrice, appelée matrice de transmission (TM), a été mesurée pour la première fois dans le (Popoff et al., 2010a). La connaissance de cette matrice permet de focaliser sur n'importe quelle cible en sortie et de reconstruire une image inconnue à travers un milieu diffusant (Popoff et al., 2010b).

La TM est l'objet mathématique utilisé pour décrire mésoscopiquement la relation linéaire existant entre les champs optiques d'entrée et de sortie du système. Elle est définie à une longueur d'onde donnée. Cette matrice $\mathbf{T}$, composée d'éléments complexes $t_{m,n}$, relie en amplitude et en phase la composante de sortie n^{th} à la composante d'entrée m^{th} (voir figure 3.9 page 92). Pour un champ d'entrée donné E^{in} , les composantes du champ de sortie E^{out} se lisent :

$$E_m^{\text{out}} = \sum_n t_{n,m} E_n^{\text{in}}, \quad (\text{D.2})$$

avec E_n^{in} et E_m^{out} respectivement l'amplitude complexe du champ optique du mode d'entrée n^{th} et du mode de sortie m^{th} . Dans un système où la lumière subit de multiples événements de diffusion, la transmission est bien représentée par une matrice aléatoire complexe (Goetschy and Stone, 2013). Dans un contexte expérimental, les bases d'entrée et de sortie pour représenter les champs doivent être judicieusement choisies pour être facilement mise en œuvre sur le SLM et efficacement détectée. La connaissance du TM pour un système temporellement stable permet de contrôler le champ optique de sortie en injectant le front d'onde d'entrée correspondant.

L'opérateur de retournement temporel L'opérateur $\mathbf{T}\mathbf{T}^\dagger$ est appelé opérateur de retournement temporel (TRO). Il s'agit d'un opérateur à matrice carrée exprimé dans la même base que l'entrée de la matrice de transmission. Cet opérateur est utilisé expérimentalement pour inverser la propagation pour une matrice de transmission mesurée. En effet, contrairement à l'inversion matricielle régulière par $\mathbf{T}^{-1}$, ce processus évite les problèmes de divergence dus à la mesure du bruit dans la matrice de transmission expérimentale (Popoff et al., 2010b).

D.3.4 Mesurer une matrice de transmission

En 2010, Popoff et al. (2010a) ont mesuré expérimentalement la TM d'une couche de peinture blanche (de nature fortement diffusant) en utilisant la modulation spatiale de la lumière. En effet, comme nous l'avons vu dans la section précédente, les techniques de mise en forme du front d'onde permettent de contrôler un nombre élevé de degrés de liberté. Expérimentalement, le TM relie les n^{th} pixels de la caméra aux m^{th} pixels

du SLM. En raison de la linéarité du système, le TM peut être mesuré dans n'importe quelle base. Comme le SLM ne permet que le contrôle de la phase, nous ne pouvons pas utiliser la base canonique du pixel du SLM. Nous avons choisi les vecteurs des matrices d'Hadamard comme base d'entrée pour sa robustesse au bruit et sa modulation à haut contraste.

Les éléments complexes de ces matrices peuvent être considérés comme des valeurs de phase binaires 0 et π , qui peuvent être facilement implémentées sur les macro-pixels du SLM (voir [figure 3.10](#) page [figure 3.10](#)). De plus, en négligeant les effets de diffraction, la puissance d'entrée globale envoyée à l'échantillon est conservée à travers les différents vecteurs de la base. Une autre propriété intéressante est que toute la surface de la base est utilisée pour chaque vecteur, ce qui maximise la puissance d'entrée et donc le rapport signal/bruit de la mesure. Un exemple de la base Hadamard et de sa mise en œuvre sur la disposition des macro-pixels SLM est présenté sur la [figure 4.2](#) page 100.

D.3.5 La matrice de transmission acousto-optique

Dans une expérience acousto-optique typique, telle que présentée dans le [chapitre 2](#), un champ optique d'entrée monochromatique à la fréquence f_L illumine un milieu inhomogène. Un faisceau ultrasonore focalisé décale la fréquence d'une partie des photons ($f_L + f_{US}$). En transmission, les photons modulés (marqués) et non modulés sont présents. La matrice de transmission acousto-optique (AOTM) représente le lien linéaire entre le champ d'entrée à la fréquence f_L et les photons marqués à la fréquence $f_L + f_{US}$ (voir [figure 3.11](#) page 95).

$$E_m^{\text{out}}(f_L + f_{US}) = \sum_n t_{m,n} E_n^{\text{in}}(f_L). \quad (\text{D.3})$$

Les photons marqués proviennent nécessairement de la région ultrasonore. Ainsi, par rapport au TM complet, l'AOTM contient des informations sur un sous-ensemble de tous les canaux possibles, ceux qui traversent la région modulée acoustiquement. La modulation acoustique n'est pas uniforme dans la zone insonifiée, tous les canaux n'apportent pas une contribution équivalente au *speckle* transmis. Comme nous l'avons vu dans le [chapitre 2](#), le profil de pression acoustique a un maximum en son centre et est bien approximé par une forme gaussienne. La forme de la sélection le long de l'axe de propagation acoustique dépend de la modulation appliquée aux ultrasons. Pour des sauts de phase aléatoires, on obtient une forme triangulaire. Pour des signaux chirpés, on obtient une fonction sincère. Dans les deux cas, l'efficacité du marquage étant proportionnelle à l'amplitude de l'onde de pression, une plus grande partie des photons passant par le centre du spot acoustique est marquée. Par conséquent, le canal passant par le centre du spot acoustique a une plus grande contribution au chatoiement enregistré par la caméra. [Katz et al. \(2019\)](#) a proposé d'analyser l'AOTM pour révéler le canal transmissif passant par le centre du spot acoustique. En effet, le guidage de la lumière à travers ces canaux offre une meilleure technique de focalisation que la technique standard d'inversion temporelle en termes de résolution. L'outil choisi pour analyser l'AOTM est l'analyse en composantes principales. Nous la présentons dans la section suivante.

D.3.6 La décomposition en valeur singulière

La décomposition en valeurs singulières (SVD) est un outil puissant en analyse matricielle. Elle consiste en une décomposition qui généralise la diagonalisation pour les cas où les espaces d'entrée et de sortie ne sont pas les mêmes. En particulier, elle fonctionne pour les matrices non rectangulaires. Elle est bien adaptée aux TM puisque le plan d'entrée et le plan de sortie sont à des positions différentes. Considérons $\mathbf{M}$ une matrice de transmission rectangulaire de forme $M \times N$. Le SVD factorise $\mathbf{M}$ en :

$$\mathbf{M} = \mathbf{U} \mathbf{S} \mathbf{V}^\dagger. \quad (\text{D.4})$$

$\mathbf{S}$ est une matrice diagonale rectangulaire de taille N avec des éléments positifs réels sur la diagonale appelés les valeurs singulières λ_i . Les valeurs singulières sont triées par ordre décroissant. Son indice est défini comme un indice de canal propre.

Dans le cas des matrices de transmission optique, les valeurs propres du TRO ($\mathbf{T} \mathbf{T}^\dagger$) correspondent au carré des valeurs singulières. Les valeurs propres de $\mathbf{T} \mathbf{T}^\dagger$ correspondent à la transmittance attendue des canaux propres correspondants. En outre, $\mathbf{U}$ et $\mathbf{V}$ sont des matrices unitaires. Leurs colonnes correspondent aux canaux propres de transmission, respectivement aux plans de sortie et d'entrée, exprimés dans les bases canoniques des pixels de la caméra et du SLM.

La SVD est un outil récurrent utilisé en ingénierie du *speckle*. Il a été utilisé pour la focalisation sélective (Popoff et al., 2011 ; Chaigne et al., 2014 ; Jeong et al., 2018), pour identifier les canaux ouverts et fermés (Vellekoop and Mosk, 2008 ; Kim et al., 2012 ; Yu et al., 2013) et de trouver des états sans dispersion à partir de la version spectrale du TM (Xiong et al., 2016 ; Ambichl et al., 2017). Dans la prochaine section, nous verrons comment la SVD de du TRO acousto-optique permet la focalisation à travers une couche diffusante.

Dans cette section, nous avons passé en revue les manipulations de front d'onde mises en œuvre au cours des deux dernières décennies. Nous avons vu que l'inversion temporelle du champ optique permet la focalisation vers les émetteurs. La conjugaison de phase optique dans les cristaux photoréfractifs est une mise en œuvre monochromatique de l'inversion temporelle. Combinés à un marquage acousto-optique, ils ont permis la focalisation au sein d'un échantillon diffusant dans une zone de taille de l'ordre de la longueur d'onde des ultrasons. Ensuite, le développement de modulateurs spatiaux-lumineux numériques permet de contrôler l'illumination d'entrée en phase ou en amplitude. Ces dispositifs permettent de construire numériquement une approche matricielle des systèmes optiques en caractérisant leurs modes d'entrée et de sortie. Une fois construites, les matrices de transmission donnent accès à l'opérateur temps-retour.

Nous avons également présenté la décomposition en valeurs singulières qui, appliquée à un système optique, permet d'obtenir le front d'onde d'entrée qui maximise la transmission optique à travers l'échantillon. Dans le chapitre suivant, nous présentons le montage optique expérimental qui permet de mesurer la matrice de transmission acousto-optique. Nous verrons ensuite comment la décomposition en valeurs singulières peut être exploitée pour focaliser à l'intérieur de la zone focale acoustique.

D.4 Mesure de la matrice de transmission acousto-optique (AOTM)

Comme expliqué dans le chapitre précédent, dans un milieu diffusant dans lequel sont focalisés des ultrasons, l'AOTM relie le champ optique d'entrée au champ de sortie des photons marqués par l'effet acousto-optique. Dans ce chapitre, nous présentons le montage optique expérimental qui permet de mesurer l'AOTM. Après avoir présenté la mesure de l'AOTM avec une modulation acoustique par des sauts de phase aléatoires, nous voyons comment la SVD appliquée à l'AOTM permet de focaliser le faisceau optique à l'intérieur de la tache focale acoustique. Permettant ainsi la limite de résolution de l'imagerie acousto-optique imposée par la diffraction acoustique. Cette valeur limite correspond à une fraction de millimètre pour une excitation de quelques mégahertz, alors que la limite de la focalisation optique est la longueur d'onde optique ($< 1 \mu\text{m}$). Par conséquent, la focalisation au-delà de la limite de diffraction acoustique permettrait de résoudre plus de détails dans le tissu et de rapprocher l'imagerie acousto-optique de la norme de résolution en microscopie.

D.4.1 Mesure de l'AOTM

Dans cette première section, nous présentons le montage optique permettant de mesurer l'AOTM.

Le montage optique Le montage optique est basé sur un montage d'imagerie acousto-optique classique dans lequel on ajoute une modulation spatiale du faisceau lumineux incident au milieu diffusant à l'aide d'un SLM (voir [figure 4.1](#) page 99). Comme pour l'AOI, nous utilisons une source laser monochromatique continue dont la longueur d'onde se situe dans la fenêtre optique thérapeutique des tissus biologiques ($\lambda = 780 \text{ nm}$). La modulation acoustique est fournie par un transducteur mono-élément, une détection par holographie numérique mesure la contribution des photons marqués du champ diffusé de sortie du milieu.

Le faisceau sonde incident au milieu est étendu et collimaté afin de couvrir toute la surface du SLM. L'échantillon diffusant que nous utilisons est une lame de verre dépoli, l'onde acoustique est focalisée au centre du faisceau lumineux dans une cuve d'eau placée derrière le diffuseur.

Façonnage du front d'onde du faisceau incident Comme proposé par [Katz et al. \(2019\)](#), nous mesurons l'AOTM dans la base des vecteurs d'Hadamard. La surface du SLM offre quelques millions de pixels donnant accès à autant de degrés de liberté. Lorsque nous modulons le front d'onde, les pixels sont en fait regroupés en macro-pixels avec une géométrie choisie. Le réseau global de macro-pixels peut être choisi pour concevoir la géométrie de la modulation et également pour s'adapter au nombre de degrés de liberté souhaité. Pour implémenter le motif hexagonal au réseau de pixels du SLM, nous utilisons une bibliothèque *Python* développée par Sébastien Popoff ([Matthès et al.](#),

2019). Le SLM est contrôlé depuis un ordinateur comme un écran secondaire. Le tableau des phases à afficher, codé sur 8 bits pixel par pixel, est envoyé par l'ordinateur au contrôleur du SLM. Comme nous l'avons vu dans la section précédente, les LCOS-SLM à réflexion sont constitués de cellules à cristaux liquides prises en sandwich entre une surface réfléchissante et une électrode transparente. Cette structure a deux effets négatifs sur la lumière incidente : tout d'abord, le réseau de pixels agit comme un réseau de diffraction bidimensionnel, l'angle de diffraction est de $\approx 3.5^\circ$ à 780 nm. Deuxièmement, la surface en verre du SLM ajoute une réflexion non modulée dans la direction de la réflexion spéculaire. Pour se débarrasser de la lumière non modulée, une rampe de phase périodique le long d'une direction est superposée au motif de modulation (voir [figure 4.2](#) page 100). Ainsi, à la sortie du SLM, une fente réglable en largeur placée au niveau du plan de Fourier d'une lentille sphérique permet de sélectionner uniquement le premier ordre de diffraction, modulé par le motif.

D.4.2 La modulation acoustique

Comme nous l'avons vu dans la [section 2.1.4](#), l'utilisation de la modulation par des sauts de phase aléatoires permet d'améliorer la résolution le long de l'axe de propagation des ultrasons. Elle permet également d'améliorer le rapport signal à bruit en utilisant un temps d'intégration plus long. Pour obtenir la même résolution que [Katz et al. \(2019\)](#), nous appliquons d'abord des sauts de phase aléatoires sur une porteuse à 15 MHz. Après de multiples essais, nous ne sommes pas parvenus à obtenir un rapport signal à bruit suffisant pour retrouver correctement la phase de la contribution acousto-optique. Nous avons émis l'hypothèse que les sauts de phase aléatoires sont trop importants du point de vue spectral par rapport à la largeur de ligne du transducteur ultrasonique. De plus, afin d'éviter une détérioration due à une surchauffe, il faut respecter la limite de dissipation de puissance dans le transducteur (125 mW). Comme la longueur temporelle des sauts de phase aléatoires (2 ms) est mille fois plus longue que les impulsions utilisées par [Katz et al. \(2019\)](#), la tension d'entrée du signal a été réduite d'un facteur dix. La quantité de photons marqués souffre de cette excitation acoustique réduite et doit être compensée par le temps d'intégration plus long permis par la méthode d'auto-corrélation telle que détaillée dans [section 2.1.4](#).

Pour ces raisons, nous avons décidé de changer pour une sonde 5 MHz que nous avons caractérisée dans le [chapitre 2](#). Comme nous l'avons vu sur la [figure 2.2](#), sa zone focale est plus large que celle de la sonde 15 MHz :

$$W_{-6\text{dB}} = 1.02 \frac{c_s F}{f D}, \quad (\text{D.5})$$

avec $W_{-6\text{dB}}$ le diamètre du faisceau des ultrasons à -6dB , F la distance focale, f la fréquence et D le diamètre de la membrane. Ainsi, en utilisant cette sonde tout en gardant la même largeur des sauts, nous multiplions la zone de marquage par neuf par rapport à la zone focale de la sonde 15 MHz (voir [figure 4.3](#) page 101). En utilisant une largeur pour les sauts de phase aléatoires de 200 ns, la zone focale acousto-optique est

elliptique avec une taille de 0.3 mm le long de la colonne acoustique et une 0.75 mm FWHM dans la direction transverse.

D.4.3 Construction de l'AOTM

La matrice de transmission est mesurée en affichant les différents vecteurs de phase correspondant aux N_{SLM} vecteurs de la base d'entrée. Les images successives, exemple présenté sur la [figure 4.6](#) page 104, sont dépliées pour constituer les lignes de la matrice de transmission. La matrice correspondant à une base de modulation à 4096 éléments, c'est-à-dire impliquant 2^{12} degrés de liberté sur le SLM, est présentée sur la [figure 4.7](#) page 105.

Ensuite, nous calculons l'opérateur de retournement temporel (TRO) :

$$\mathbf{O} = \mathbf{T} \mathbf{T}^\dagger, \quad (\text{D.6})$$

de taille 4096×4096 , reliant la base canonique des pixels du SLM à elle-même par un aller-retour à travers le système (voir [figure 4.8](#) page 106).

D.4.4 Focalisation de la lumière à l'intérieur de la zone focale acoustique à l'aide de la décomposition en valeurs singulières

La décomposition en valeurs singulières d'une matrice carrée permet d'identifier les vecteurs singuliers du système et de les trier dans l'ordre décroissant de leurs valeurs singulières correspondantes. Appliqués au TRO acousto-optique, les vecteurs singuliers associés aux valeurs singulières les plus élevées représentent les vecteurs complexes, exprimés dans la base canonique des segments sur le SLM, qui conduisent le plus grand nombre de photons à marquer à travers le foyer ultrasonore. Inversement, les derniers vecteurs de phase, correspondant à la valeur singulière la plus faible, fournissent les champs optiques à travers lesquels l'interaction acousto-optique est la plus faible.

Une fois que nous avons les vecteurs singuliers, nous pouvons les propager virtuellement à travers le système en utilisant l'AOTM mesurée en calculant le champ de sortie $E_{\text{out}} = \mathbf{T} E_{\text{in}}$. Ainsi, nous calculons le champ acousto-optique qu'ils produiraient s'ils étaient affichés par le SLM. De plus, comme la propagation de la lumière du plan des ultrasons au plan de la caméra se fait en espace libre, nous récupérons le champ dans le plan des ultrasons directement en traitant une transformée de Fourier bidimensionnelle inverse. [figure 4.10](#) et [figure 4.11](#) page 107 présentent le champ complexe calculé dans le plan ultrasonore correspondant aux vecteurs singuliers d'indices croissants. L'efficacité du marquage augmentant avec l'amplitude locale dans la zone focale acoustique, les premiers vecteurs singuliers correspondent à des fronts d'onde qui focalisent la lumière au centre de la tache acoustique. En effet, à cet endroit, l'efficacité du marquage ultrasonore est maximale. Les vecteurs singuliers suivants correspondent à des champs optiques dans le plan objet qui couvrent une plus grande surface autour du point central du premier vecteur singulier. À partir de l'indice 100, l'intensité de l'illumination forme un motif en forme d'anneau (voir [figure 4.11](#) page 108). Ces formes également observées par [Katz et al. \(2019\)](#), correspondent au contour de la fonction de marquage à une amplitude

donnée. Cet effet débute à un indice dépendant du rapport entre le nombre de grains de *speckle* à l'intérieur de la zone focale ultrasonore et le nombre de degrés de liberté fournis par la modulation du SLM. [Katz et al. \(2019\)](#) observent les premiers anneaux à partir de l'indice 10. Dans notre cas, ils apparaissent autour de l'indice 100 pour une base de 4096 éléments. Cette différence pourrait s'expliquer par les différentes zones focales ultrasonores entre le 15 MHz utilisé dans ([Katz et al., 2019](#)) et le 5 MHz que nous utilisons ici. En effet, le nombre de degrés de liberté nécessaires pour traiter des modes distincts à l'intérieur du spot acoustique croît avec le nombre total de grains de *speckle* à traiter dans le volume étiqueté.

Nous remarquons également que nos anneaux sont plus étroits le long de l'axe acoustique. Cette observation est compatible avec l'anisotropie de notre zone de sélection due à l'utilisation de sauts de phase aléatoires. En effet, nous utilisons des sauts de phase d'une durée de $0.67 \mu\text{s}$, correspondant à une résolution axiale de $100 \mu\text{m}$, tandis que la résolution transversale est fixée par la largeur de tache focale ultrasonore de $100 \mu\text{m}$ à 15 MHz. Enfin, la résolution de la focalisation doit être comparée à l'efficacité de la méthode TRUE [Liu et al. \(2015\)](#). Cette méthode consistant à complexer par conjugaison un champ acousto-optique aléatoire, la zone focalisée est donc étalée sur la zone de marquage, soit 0.75 mm . La [figure 4.12\(a\)](#) page [figure 4.12](#) montre l'intensité de l'illumination focalisée produite par l'injection du premier vecteur singulier. Le FWHM transversal de cette illumination focalisée est de $\approx 180 \mu\text{m}$. L'augmentation de la résolution est similaire à celle obtenue par [Katz et al. \(2019\)](#). Nous avons alors focalisé la lumière sur un point 4 fois plus petit que la dimension de la tache focale ultrasonore. De plus, la résolution obtenue surpasse celles obtenues par [Ruan et al. \(2015\)](#) avec la focalisation de la lumière iTRUE. La décomposition en valeurs singulières fournit directement l'illumination optimale pour la focalisation au centre de la zone focale acoustique, ce qui est réalisable pour iTRUE après un nombre infini d'itérations. Cela démontre que la focalisation et l'imagerie acousto-optique ne sont pas fondamentalement limitées par la longueur d'onde de l'excitation ultrasonore.

La [figure 4.12](#) page [110](#) compare la focalisation expérimentale, mesurée par une caméra d'observation conjuguée au plan ultrasonore, avec le champ acousto-optique, récupéré en calculant la transformée de Fourier inverse sur le champ mesuré par la détection par holographie. On observe que la projection numérique du premier vecteur singulier (voir [figure 4.12\(a\)](#) page [110](#)) correspond au champ réel mesuré par la caméra d'observation. La largeur de la tache focale optique est la même pour les deux images, ce qui prouve l'efficacité de la mise au point. Nous produisons la même projection pour le cinquantième vecteur singulier sur la [figure 4.13](#) (page [110](#)) avec la même résolution. Cependant, la forme annulaire des vecteurs d'ordre supérieur n'apparaît pas. Cela pourrait être dû à des erreurs de mesure de l'AOTM dues à la décorrélation du milieu. En raison du long temps de mesure, les vecteurs singuliers injectés après la mesure conduiraient à des résultats différents de ceux calculés, car le milieu a changé au fil du temps.

Pour conclure cette partie expérimentale de la mesure de l'AOTM, nous avons vu comment nous mesurons l'AOTM. Fondé sur la détection hors-axe des photons marqués acousto-optiquement, le dispositif expérimental nécessite une synchronisation et une cor-

respondance de phase stables sur plusieurs heures. Ensuite, en utilisant la décomposition en valeurs singulières appliquée à l'opérateur de retournement temporel, méthode précédemment prouvée par [Katz et al. \(2019\)](#), nous nous concentrons à travers une couche de diffusion au-delà de la limite de diffraction acoustique imposée par l'excitation ultrasonique. Ainsi, nous atteignons une résolution 4 fois plus étroite que les méthodes d'étoiles guides existantes basées sur le couplage acousto-optique, comme la conjugaison de phase TRUE. De plus, nous atteignons la limite de focalisation asymptotique de la méthode itérative TRUE (iTRUE). En effet, le premier vecteur singulier s'adresse aux quelques grains de *speckle* optique situés au centre de la zone focale acoustique. Les vecteurs singuliers d'indice supérieur s'adressent ensuite aux grains de *speckle* périphériques équivalents, créant des formes d'anneaux, suivant la géométrie de la zone focale acoustique. L'adressage sélectif des grains de chatoiement suivant le profil d'excitation des ultrasons est très prometteur. De plus, nous conservons ici l'excitation du transducteur à un seul élément qui contraint géométriquement la forme de la fonction de marquage acousto-optique. Dans des travaux futurs, nous pourrions imaginer d'utiliser une sonde acoustique multi-éléments telle que la sonde échographique utilisée dans le [chapitre 2](#) pour fournir une interaction acousto-optique plus complexe.

Conclusion

Dans cette thèse, nous avons développé des approches matricielles en imagerie acousto-optique. Ces approches reposent sur la mise en forme du front d'onde des ondes acoustiques et optiques.

Après avoir rappelé l'importance des techniques d'imagerie optique pour le diagnostic médical et la détection précoce, le premier chapitre a mis en évidence les limites des techniques d'imagerie optique standard dans les tissus biologiques en raison de multiples événements de diffusion. Néanmoins, nous avons montré que la tomographie optique diffuse permet de récupérer la carte d'absorption et de diffusion à l'intérieur des milieux en résolvant le problème inverse de la propagation. Mais cette méthode entièrement optique exige des ressources informatiques élevées et la recherche d'un compromis entre la profondeur d'imagerie et le rapport signal/bruit. Par conséquent, les techniques multi-ondes tirent parti de la propagation balistique des ondes acoustiques pour sonder le contraste optique en profondeur. L'effet acousto-optique permet de marquer la lumière passant dans une région spécifique et ainsi de récupérer une résolution acoustique-diffraction à des profondeurs de l'ordre de quelques centimètres. L'amplitude de la lumière marquée est directement liée au contraste local dû à l'absorption et à la diffusion. Le brûlage de trous spectral relève le défi de la détection de la lumière marquée, tant en réflexion qu'en détection, et se rapproche des applications *in vivo*. La méthode de détection que nous utilisons, l'holographie numérique hors axe, permet de récupérer le champ optique complexe qui est un élément clé pour suivre la phase de l'onde diffusée.

Le deuxième chapitre présente différentes stratégies de modulation qui ont été développées pour l'imagerie acousto-optique. La modulation temporelle des ultrasons focalisés spatialement permet de sélectionner une profondeur plus étroite que la zone de focalisation de la sonde. Ensuite, les réseaux de transducteurs permettent des excitations spatialement larges, améliorant le rapport signal/bruit. Cette amélioration se traduit par un temps d'acquisition plus rapide compatible avec les sessions d'imagerie dans des échantillons dynamiques tels que les tissus *in vivo*. En outre, la structuration spatiale et temporelle de l'excitation acoustique permet d'obtenir une imagerie par transformée de Fourier. Cette méthode consiste à adresser les composantes spatiales de Fourier du plan d'illumination en utilisant un motif périodique bidimensionnel joué par la sonde d'échographie. Les simulations sur les distorsions acoustiques mettent en évidence un doublement de fréquence dans le domaine de Fourier qui limite néanmoins les excitations structurées possibles. Ces effets de diffraction pourraient être considérés comme des avantages de l'excitation multi-spectrale en mesurant les fonctions de marquage pour chaque motif dans un environnement connu, par exemple l'observation balistique dans un réservoir d'eau. Nous pourrions ensuite adapter la reconstruction en tenant compte de tout le spectre de Fourier bidimensionnel du motif de marquage, y compris les effets des bords du motif et de la diffraction acoustique.

Les chapitres suivants se sont concentrés sur le contrôle du front d'onde de l'illumination d'entrée. Dans le troisième chapitre, nous avons présenté les techniques de mise en forme du front d'onde et leur efficacité pour compenser la distorsion de phase appliquée

par la diffusion. La mesure de la matrice de transmission acousto-optique fournit une information globale sur les modes optiques transmis à travers le foyer ultrasonore. La décomposition en valeurs singulières que nous avons réalisée permet de mettre en évidence les modes d'entrée les plus efficaces, identifiés comme les vecteurs propres de l'opérateur d'inversion temporelle. La modulation spatiale de la lumière permet de mettre en phase le front d'onde d'entrée pour traiter ces modes. Par conséquent, le premier vecteur singulier de l'opérateur d'inversion temporelle guide la lumière au centre de la zone focale des ultrasons. Cette projection permet de focaliser à travers une couche diffusante avec une localisation plus étroite d'un facteur 4 par rapport aux techniques de conjugaison de phase optique. De plus, l'adressage sélectif des grains de *speckle* suivant le profil d'excitation des ultrasons est prometteur.

Ce travail suggère quelques axes d'amélioration. L'utilisation de l'excitation par ondes planes structurées ultrasonores pourrait bénéficier de la mesure de la fonction de marquage. Ces mesures permettraient de construire une matrice des composantes spectrales de chaque motif structurant dans le domaine spatial de Fourier. L'inversion de cette matrice permettrait d'améliorer la reconstruction en prenant en compte l'ensemble des composantes spectrales de l'excitation. De plus, nous avons vu que l'utilisation de toute la surface du capteur de la caméra améliorerait la sensibilité d'un facteur 16 par rapport aux résultats présentés dans le [chapitre 2](#). Néanmoins, cette amélioration nécessite d'optimiser le traitement de la reconstruction de l'image en passant à des unités de traitement graphique.

L'utilisation d'une barrette ultrasonore serait une grande amélioration pour la mesure de la matrice de transmission acousto-optique. En effet, le réseau de transducteurs offre une plus grande ouverture numérique, ce qui signifie qu'il produit un foyer ultrasonore plus intense pour amplifier le signal du photon marqué. De plus, l'indépendance des canaux de transduction de ces sondes permet des foyers multiples, ce qui permet de mesurer des AOTM plus complexes. L'utilisation d'un appareil d'échographie demande néanmoins d'adapter l'architecture de déclenchement afin de synchroniser les excitations ultrasonores avec la caméra. Ensuite, le temps d'acquisition de l'AOTM est actuellement dominé par le déchargement de la mémoire de la caméra. De nombreuses alternatives matérielles et logicielles ont été testées au cours de la thèse afin de réduire cette durée. Une caméra permettant une visualisation en continu tout en déclenchant des enregistrements devrait constituer une amélioration significative.

Enfin, comme présenté dans [chapitre 2](#), la modulation par des harmoniques d'ordre supérieures a été testée avec un transducteur à élément unique mais pas encore avec une sonde d'échographie. Comme les appareils d'échographie permettent de générer des formes d'onde arbitraires indépendamment sur chacune de ses voies, des sauts de phase aléatoires en demi-phase pourraient être mis en œuvre sur ceux-ci pour bénéficier de la PSF plus nette de la seconde harmonique.

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RÉSUMÉ

L'imagerie est un outil clef du diagnostic médical. Les méthodes d'imagerie optique classiques telles que la microscopie ou la tomographie de cohérence optique se heurtent à la diffusion multiple de la lumière au bout de quelques libres parcours moyens de transport ($\sim$ mm) dans les milieux biologiques. Fondée sur la détection de photons issus d'une source de lumière cohérente et marqués par un champ acoustique, l'imagerie acousto-optique est une technique d'imagerie optique en milieux fortement diffusants avec une résolution millimétrique. Pour cela les techniques de façonnage de l'onde acoustique permettent de maîtriser la zone dans laquelle le marquage de la lumière par les ultrasons a lieu. La quantité de photons marqués étant proportionnelle à l'intensité lumineuse locale de la zone insonnifiée, la sélection de la lumière marquée en sortie du milieu permet ainsi de remonter à une information de contraste optique dans la zone couverte par les ultrasons. Nous pouvons alors reconstruire la carte d'intensité à l'intérieur du milieu diffusant. Dans ce travail, nous utilisons l'holographie hétérodyne hors-axe comme méthode de détection des photons marqués. Cette méthode interférométrique utilise une onde de référence à la fréquence des photons marqués. Les franges d'interférences sont échantillonnées par les pixels de la caméra ce qui permet la reconstruction sélective du champ lumineux. Des résultats expérimentaux sont présentés accompagnés de simulations. Nous développons l'illumination acoustique en ondes planes structurées. Cette méthode permet d'imager un plan en sondant ses composantes de Fourier. Combinée avec la détection sur caméra, elle permet de proposer un temps d'imagerie compatible avec le contexte de l'*in vivo*. Nous avons ensuite investigué des modulations acoustiques non-linéaires de la lumière pour réduire la zone de marquage. Enfin, les techniques de façonnage du front d'onde optique nous ont permis de mesurer la matrice de transmission acousto-optique du milieu. Cette matrice lie le front d'onde optique entrant au champ marqué sortant. Nous l'avons mise à profit pour mettre en évidence les canaux de transmission contraints au travers de la tache acoustique. Nous montrons que l'étude de cette matrice par analyse en composantes principales permet de focaliser l'onde optique à l'intérieur de la tache acoustique et ainsi de vaincre la limite de résolution acoustique.

MOTS CLÉS

Acousto-optique, acoustique, optique, milieux diffusant, holographie hétérodyne hors-axe, imagerie par transformée de Fourier, modulation acoustique non-linéaire, façonnage du front d'onde

ABSTRACT

Imaging is a core element of medical diagnosis. Observation depths of standard optical imaging techniques, such as microscopy or optical coherence tomography, are limited by multiple scattering to few transport mean free paths ($\sim$ mm) in biological tissues. Acousto-optic imaging is an optical imaging technique in highly scattering media based on the detection of photons from a coherent light source tagged by an acoustic field. Acoustic wave shaping techniques allow to control the area in which this ultrasonic light marking takes place. The tagged-photon quantity is proportional to the local light intensity in the insonified area. Selecting the tagged light at the output of the medium allows retrieving the optical contrast in the area covered by the ultrasounds. We can then reconstruct the intensity map inside the scattering medium. In this work we use off-axis heterodyne holography to detect the tagged photons. This interferometric method uses a reference wave at the frequency of tagged photons. Interference fringes are sampled by the pixels of the camera. Successive images are combined to reconstruct the field of the tagged light. Experimental results are presented along with simulations. We then develop structured plane wave illumination. This method allows imaging a plane by probing its Fourier components. Combined with camera-based detection it offers an imaging duration compatible with *in vivo* implementations. We then investigate non-linear acoustic modulation of light to reduce the tagging area. Finally, optical wavefront shaping techniques provide a measure of the acousto-optic transmission matrix of the media. This matrix links the input optical field to the output tagged field. Once measured, we use it to retrieve the transmissive channels constrained through the acoustic spot. We study this matrix by principal component analysis to allow focusing the optical waves inside the acoustic spot, thus overcoming the acoustic resolution limit.

KEYWORDS

Acousto-optic, acoustics, optics, imaging, scattering media, off-axis heterodyne holography, Fourier-transform imaging, non-linear acoustic modulation, wavefront shaping