Ultra-broadband Dielectric Beam Steering Mirrors for *in vivo* Nonlinear Microscopy: How Mirror Performance Affects Imaging Quality and Laser Safety Issues?

Tímea Grósz[1,2], Attila P. Kovács[1], Róbert Szipőcs[2,3,*]
[1]*Department of Optics and Quantum Electronics, University of Szeged, Dóm tér 9, H-6720 Szeged, Hungary*
[2] *R&D Ultrafast Lasers Ltd, P.O. Box 622, H-1539 Budapest, Hungary*
[3]*Wigner RCP, Institute for Solid State Physics and Optics, P.O. Box 49, H-1525 Budapest, Hungary*
* *r.szipocs@szipocs.com*

Abstract: Broadband dielectric beam steering mirrors are key components for nonlinear microscopes comprising tunable Ti:sapphire lasers. We characterize dispersion of a few mirror samples by spectral interferometry and show how it affects imaging quality.
OCIS codes: (310.1620) Interference coatings; (320.0320) Ultrafast optics; (180.4315) Nonlinear microscopy.

1. Introduction

Nonlinear microscopy is being increasingly used to perform *in vivo* studies in life sciences, for instance in dermatology, neurology or pharmacology [1,2]. The combination of different modalities such as two-photon absorption fluorescence (TPAF), second harmonic generation (SHG) or coherent anti-Stokes Raman spectroscopy (CARS) are promising techniques to obtain detailed morphological and structural information about living tissues by the use of femtosecond lasers operating in the red, near-infrared spectral range (680-1300 nm). In dermatology, the ability to follow the changes and distribution of various biological compounds in the skin is essential for better understanding the mechanism of diseases. Nonlinear microscopy is a noninvasive, high-resolution and deeply penetrating optical imaging technique with high sensitivity. The epidermis and dermis both contain numerous endogenous chromophores, such as NADH, melanin, keratin, elastin and collagen that can be visualized by various excitation wavelengths, without the need of exogenous contrast agents, which condition is required for *in vivo* investigations.

In order to minimize the thermal load of living tissues, the excitation wavelength has to be properly set for the different chromophores while keeping the pulse duration close to the transform limit, i.e. phase distortion of the optical pulses has to be kept as low as possible between the tunable, femtosecond pulse laser source (typically a Ti:sapphire laser) and the biological sample.

Broadly tunable Ti:sapphire lasers comprise ultra-broadband chirped dielectric mirrors [3] for broadband feedback over the full gain bandwidth of Ti:sapphire lasers, which practically ranges from ~680 to ~1040 nm. Beside the high reflectance (R > 99.8%), these dielectric mirror coatings exhibit a smooth, monotonous variation of group delay vs. wavelength function over the high reflectance band supporting sub-100-fs performance all over the tuning range [3].

In contrast to (femtosecond pulse) laser oscillators, where the (ultra-broadband chirped) mirrors are used at near normal incidence, nonlinear microscopes require several low-absorption loss, broadband beam steering dielectric mirrors for 45°angle of incidence (AOI): a few of them between the laser source and the microscope, and at least two of them in the laser scanner unit. The high reflectance bandwidth of these beam steering mirrors should match the full tuning range of the laser source for both s- and p-polarized light, while the phase distortion upon reflection has to be minimized for both polarization states (e.g., in the scanner unit). We must mention that the use of protected silver mirrors is not advised, since thermal effects may strongly reduce the focusability of laser beam, hence the quality of imaging.

In our study in 1997 [4], we found that the group-delay vs. wavelength function of ultra-broadband chirped dielectric mirrors exhibit strong resonances when these mirrors are used for s-polarized light at 45°AOI, that might strongly distort sub-100-fs optical pulses after a few reflections on these mirrors.

In our present work we investigated dispersive properties of some commercial ultrabroadband dielectric mirrors being used in commercial nonlinear microscope systems, such as in a LSM 7MP microscope (Carl Zeiss, Germany). We applied spectrally resolved white light interferometry for phase characterization of our selected mirror samples [5,6].

2. Experimental setup

The broadband dielectric mirrors under test were investigated using spectrally resolved white light interferometry [5,6]. The experimental setup depicted in Fig. 1 is a combination of a Michelson interferometer and a spectrometer (Avantes 3648, 200-1100 nm, spectral resolution: 1 nm). The interferometer was illuminated with a fiber-coupled tungsten halogen lamp (Ocean Optics HL-2000). The light was collimated by a microscope objective (Olympus Plan N 10x, NA=0.25) and then sent through a linear polarizer (Thorlabs, LPNIR050-MP Linear Polarizer, 650 - 2000 nm) towards the beam splitter. The area marked with dashed lines denotes the adjustable part of the reference arm. Each mirror under test was placed in the sample arm of the interferometer at 45° AOI. The other mirrors in the setup were protected metallic mirrors. Spectrally resolved interferograms were recorded for both s- and p-polarized light by adjusting the polarizer. The reference arm length was set to get dense fringes permitting the use of the Fourier-transform evaluation method, the applicability and precision of which has already been demonstrated in measuring dispersion of specialty fibers and other optical components as well. Additionally, this method offers the best results when the group delay vs. wavelength functions exhibit some resonant features [6].

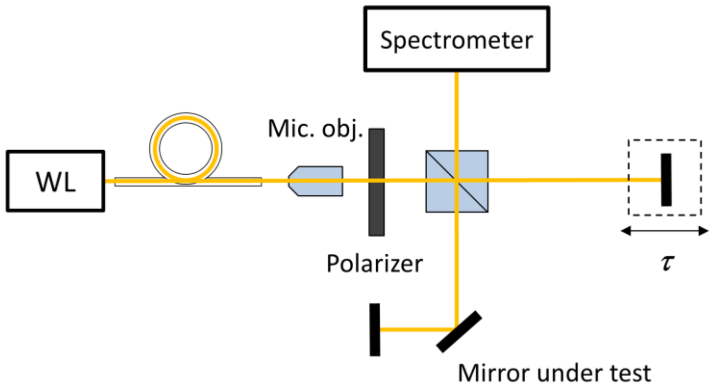


Fig. 1 Experimental setup.

3. Results

During our investigations we tested several commercially available ultra-broadband dielectric mirrors, some of them being used as beam steering mirrors in nonlinear microscope systems, such as the LSM 7MP microscope of Carl Zeiss (Jena, Germany). For example, Carl Zeiss uses Code No. 508-519 ultrabroadban dielectric mirrors (Newport Corp.). In other microscope setups, users often use the Code No. BB1-E03 broadband dielectric mirrors (Thorlabs Inc.) for beam steering between the tunable femtosecond pulse laser source and the microscope. For comparative purposes, we also tested some of our ultrabroadband chirped mirrors developed for intra-cavity applications in Ti:sapphire lasers. In Fig. 2, the measured group delay vs. wavelength functions of the ultra-broadband dielectric mirrors of Newport Corp. (Code: 508-519) and Thorlabs Inc. (Code: BB1-E03) are shown.

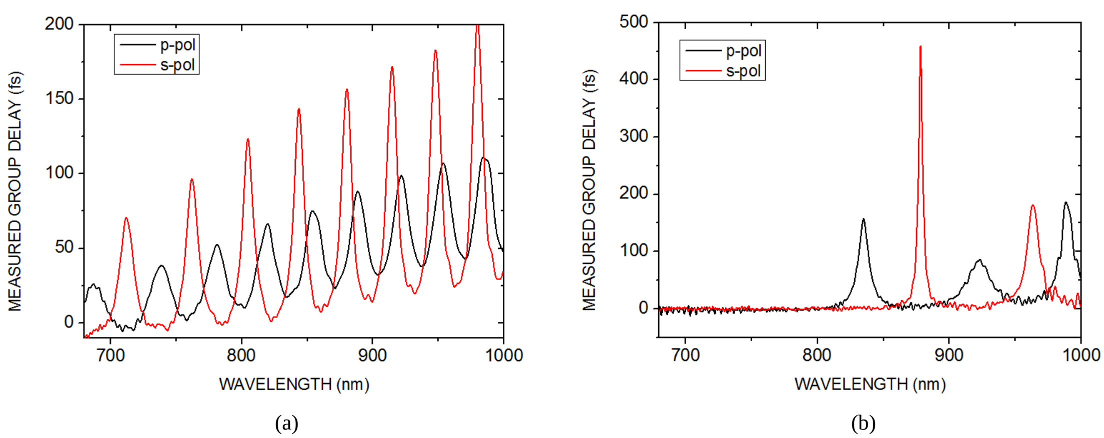
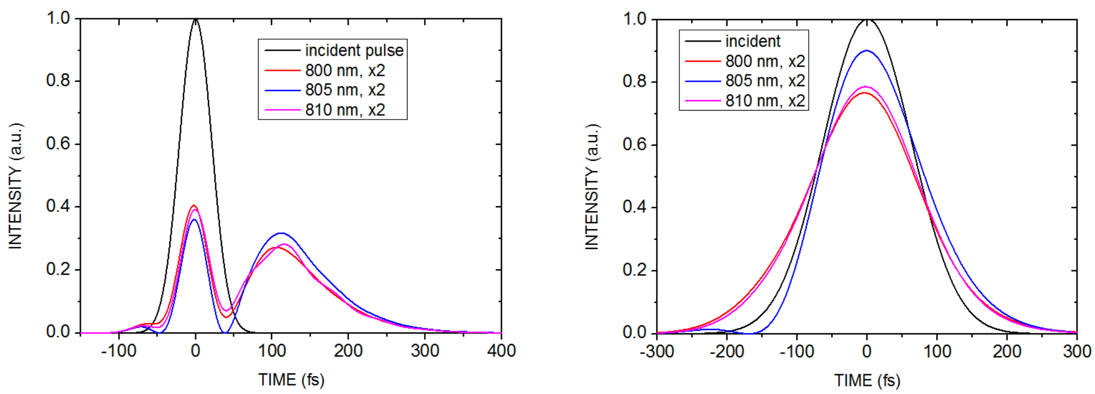


Fig. 2 Measured group delay vs. wavelength functions of ultra-broadband dielectric mirrors (a) CODE: 508-519 (Newport Corp.) and (b) BB1-E03 (Thorlabs Inc.)

One can observe that the Newport mirror exhibit a large number resonances for both p- and s-polarized light, however, for s-polarized light the maximum value of group delay is considerably higher and the resonance is much narrower, which results in much higher group delay dispersion (GDD) values. In the case of the mirror sample from Thorlabs Inc, the number of resonances is much lower over the high reflectivity range, but the value of group delay is extremely high at around 880 nm for s-polarized light. Such a resonance may well increase the pulse duration from 0.1 ps to 1 ps after a few bounces on these mirrors.

In Fig. 3 we show the computed intensity vs. time functions of transform limited (a) 50 fs and (b) 150 fs pulses being reflected on the ultra-broadband dielectric mirror 508-519 (Newport Corp.) for s-polarized light. We note here that these mirrors are used in a commercial laser scanning two-photon excitation fluorescence microscope system of Carl Zeiss, for which a Mai Tai laser (product of Newport- Spectra Physics) is used as a tunable laser source. The laser delivers nearly transform limited laser pulses of ~50 fs. In Fig. 3(a) and Fig. 3(b), one can observe how the temporal shape of the optical pulse is changed after two mirror reflections for a 50 fs and a 150 fs pulses, respectively. For the 50 fs pulse, the peak intensity is reduced by a factor of ~2.5, which results in a similar reduction in the fluorescence signal in the microscope. It can be compensated by using a higher laser power, but it often results in damage of the biological sample. If one uses longer pulses [7], the mirrors do not have such an drastic effect: the peak intensity is reduced to 75-85 % of the original value depending on the central wavelength of the pulse (see Fig. 3(b)).



(a) (b)

Fig. 3 Calculated intensity vs. time functions of transform limited (a) 50 fs and (b) 150 fs pulses being reflected on an ultra-broadband dielectric mirror 508-519 (Newport Corp.) for s-polarized light at 45° AOI.

Summarizing our result we can say that imaging quality of nonlinear microscope systems strongly depends on dispersive properties of broadband dielectric beam steering mirrors. Unfortunately, current commercial mirrors have limited performance regarding dispersion, which might require different design approaches in the near future.

4. References

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