Simple system for simultaneous in situ MEFISTO pulse characterization of broadband pulses and high resolution nonlinear imaging

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Multiphoton microscopy had been developed as a powerful technique for high resolution fluorescence imaging due to the development of broadband ultrashort light pulses [1]. The capability of a simple setup to obtain broadband pulses which are reproducible is an important advantage in biological applications. Here we generate controlled broadband pulses using the output pulse of a Ti:Sapphire laser ($\Delta T \sim 180$ fs and spectral width $\Delta \lambda \sim 10$ nm) traveling through insensitive bending single mode fiber (OFS Micro820-16 Fiber) with a length of L=10cm. A pair of prisms (SF10) is used to control the chirp of the pulses from the fibre. The pulses are then sent to an Adapte inverted Nikon TE2000–U microscope and are fully characterized, at the sample plane of the microscope using the MEFISTO technique [2]. The uncompensated pulses showed a temporal duration of $\Delta T \sim 875$ fs and spectral width $\Delta \lambda \sim 50$ nm and a coupled power of $P \sim 200$ mW. Numerical results obtained using a simple model agreed with experimental results. The stability and the shape of the spectrum of the output pulse was controlled by the coupled power in an easy way. By compensating the chirp of the pulse using the pair of prisms apex separation 94 cm, it was possible to compensate for the quadratic dispersion introduced by the system [the fiber and the microscope (NA=1.4)]. The compressed pulses were $\Delta T \sim 35$ fs duration (FWHM). We imaged a suspension of green fluorescent polystyrene microspheres using two photon excited fluorescence (TPEF) in the backward direction (Fig. 2 (a)). At the same time we collected the second harmonic generation (SHG) signal from starch granules in forward direction (Fig. 2(b)). This SHG signal was also used for in situ characterization of the pulses.

As a conclusion, we have proposed a simple technique to increase the available bandwidth of the Ti:Sapphire pulses. We have shown that it is possible to simultaneously characterize the pulses at the sample plane of a nonlinear microscope during imaging the studied sample. This simple set up can be used for more complex imaging where pulse shaping could be needed.