High-Resolution Broadly-Tunable MOPA-Based Terahertz Spectrometer to Non-Destructively Probe and Modulate Protein Electrodynamics

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Abstract

We report on the development of a high-resolution (< 1 GHz at 1.0 THz), broadly–tunable (~0.5 - 3.0 THz) fiber MOPA-based terahertz spectrometer system. The flexibility and performance of this system will be demonstrated with gaseous (water vapor) and aqueous (water and solvated protein) samples with a view towards selectively and non-destructively probing functionally significant large-scale protein motions.

Introduction and Background

A narrow-bandwidth terahertz spectrometer with a tuning range limited to 0.4 THz from a central wavelength has been previously demonstrated in our group by Malinowski et al.[1]. In this work we demonstrate our progress towards a significantly upgraded system tunable over a range in excess of 2.0 THz (Fig.1).

Since protein folding dynamics take place on the picosecond timescale, vibrational modes tend to lie in the sub-2THz frequency range[2,3]. Many recent studies have made extensive use of time-domain THz spectroscopy (TDS)[4,5]. The limited spectral resolution of the technique has generated a clear need for high-resolution THz sources, and recent development in this regard has been rapid and widespread[6,7,8].

Our THz system has a similar dual fiber master oscillator power amplifiers (MOPA) chain design producing two pulsed beams of 2 ns duration at a repetition rate of 0.5 MHz. The maximum peak power per channel is ~5 kW (5 W average power).

Both MOPA chains are designed to minimize spectral linewidth: one amplifier chain is based on a custom-built distributed feedback (DFB) laser operating at ~1060 nm, and the other is based on a Toptica DL pro 1040 tunable external cavity laser with a tuning range of 980 - 1075 nm. The linewidth before amplification for both chains is of the order of 100 kHz.

THz radiation is generated via difference frequency generation (DFG) in a (110) cleaved GaP crystal which is directed through a custom-built fluidic cell mounted in front of a silicon filter and pyroelectric THz detection system. A half-wave plate is located between the main system lens and the GaP crystal to achieve optimum polarization orientation with respect to the crystal orientation.

Both MOPA chains exhibit a flat gain response over the entire tuning range, with an optical signal-to-noise ratio (OSNR) of ~30 dB (as measured at 0.1nm resolution) which is particularly flat in the narrow range corresponding to the low frequency (0.5 - 1.0 THz) region in the DFG process (Fig. 2). The estimated spectral linewidth of the MOPA output is ~500 MHz at 1060nm, corresponding to a ~1 GHz effective linewidth at 1.0 THz with a typical peak THz power of ~ 3 mW.

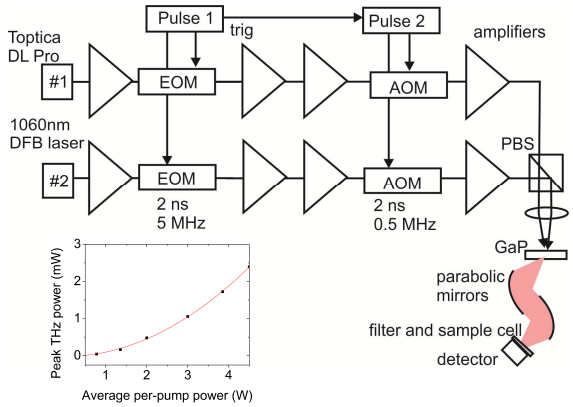

Fig. 1. ORC THz spectrometer design, showing dual fiber MOPA with PBS for beam combining and angle-tuning. Inset: THz power as a function of average incident pump power per MOPA chain.

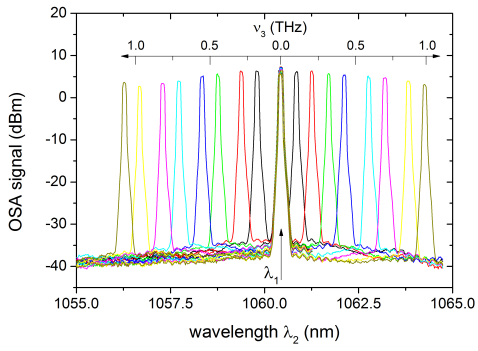

Fig. 2. MOPA output spectra. One MOPA chain is fixed at λ_1 ~ 1060 nm, the other chain (λ_2) is tunable. OSA resolution = 0.1 nm.

DFG of THz radiation can be achieved by passing both beams through the GaP crystal in either a collinear or (as in our case) in a non-collinear phase-matched configuration[1]. The tunable beam passes through the GaP perpendicular to the crystal face whilst the fixed wavelength (1060nm) beam is passed through at an angle adjustable by translating the polarizing beam splitter (PBS) which is mounted on a micrometer-driven translation stage. By translating the PBS along the axis perpendicular to the GaP crystal face, the fixed wavelength beam's path is displaced perpendicularly to the axis of propagation and parallel to the optical table. The center of the GaP crystal sits at the shared focal point of both beams, translating the PBS corresponds to angle tuning the two beams. The Rayleigh ranges of the two beams are much longer than the thickness of the GaP crystal, ensuring efficient beam overlap.

To facilitate high-resolution vibrational spectroscopy of proteins the following upgrades have been implemented:

(i) Overall control of the THz spectrometer is consolidated in a single standard PC running Windows XP. An 8-port RS-232 card and USB system is used to communicate with the various computer-controllable components of the system.

(ii) The entire THz generation and detection sections are contained within a Perspex N_2 purging shroud which features wedged antireflection coated entrance windows to eliminate reflections and etalon effects, and electrical feed-throughs.

(iii) Our compression-sealed flow cell for fluidic THz characterisation and protein manipulation is similar to a previous design[9,10]. Polytetrafluoroethylene (PTFE) film is compressed between two 5 mm thick PTFE discs by a 3D printed manifold, where a cutaway section of the film defines the microfluidic channels. The flow cell is mounted normal to the THz beam so the film thickness defines the path length of the biological sample. Holes are drilled in the lower manifold to connect capillaries to the microfluidic channels, allowing the delivery and disposal of multiple reagents as required for each protein experiment. .

(iv) A simplex lock-in amplifier type pyroelectric detector (Gentec T-Rad-USB) with an integration time of approximately 5s is used to measure the throughput THz power. Using our narrow-band source, we aim to selectively probe and excite functionally relevant protein vibrational modes without denaturing the molecule. Such vibrational modes have previously only been non-specifically excited with a broadband source to cause denaturation[11]. Of particular interest are the lower frequency modes in the sub-1.0THz region[12], as these modes are most likely to be associated with functionally significant large-scale protein motions.

Results

To demonstrate the suitability of our fiber laser driven DFG based approach for precision spectroscopic measurements, we show early water vapor absorption data (Fig 3) comparing the DFG approach with conventional TDS with excellent agreement. We are soon to embark on novel studies in THz-induced protein dynamics, the confirmation of which would have significant implications for potential therapeutic applications. We hope to present our high-resolution vapor and liquid-phase spectroscopic results in the very near future.

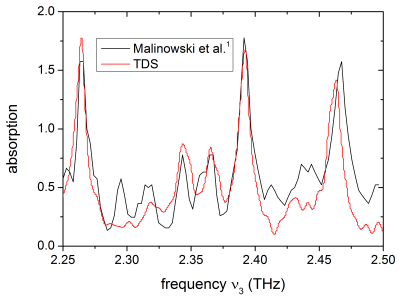

Fig. 3. Comparison of TDS and ORC system performance; water vapor absorption spectra in air are shown1.

Outlook

The terahertz spectrometer system described is capable of high-frequency-resolution excitation of protein folding, which will be exploited to selectively modulate the structure and function of proteins. Provision has been made for precise electronic control of exposure conditions in a flow-cell with transparent windows at THz and visible frequencies. Functional changes of proteins exposed to THz radiation will be evaluated using a well-characterized protein function test in free solution in the cell. Measurements on monomolecular films of protein immobilized at dielectric surfaces, equivalent to high sensitivity (multiple incidence) ellipsometry, have been demonstrated for calmodulin using commercial dual-polarization waveguide interferometry[13] and similar complementary measurements here, under THz irradiation, would further elucidate observed protein function changes. While THz-induced changes in protein dynamics may not necessarily result in conformational changes, the demonstration of such an effect, especially when combined with assessments of protein function, would lend strong support for the protein electrodynamics hypothesis postulating functionally-specific interaction of THz radiation with proteins.

Acknowledgement

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