Title: **Ultrafast Raman Spectroscopy. Construction of the setup and application to selected chemical systems.**

Author: **Michał Nejbauer**

Supervisor: **prof. Czesław Radzewicz**

Abstract

The subject of the thesis is the construction of a system for femtosecond stimulated Raman spectroscopy (FSRS) and the demonstration of its capabilities on a number of selected chemical and biochemical systems. FSRS is a relatively new technique that acquires Raman spectra of molecules by recording broadband spectrum of stimulated Raman scattering with use of a multi-picosecond narrow-bandwidth Raman pulse and a broadband probe pulse. Combined with the third, also ultrashort, light pulse FSRS allows one to record transients Raman spectra with femtosecond time resolution.

The system constructed within this project is based on a commercial Yb-doped laser emitting 440 μJ pulses at 1030 nm with 180 fs pulse duration at 1kHz repetition rate. To generate three pulses with specific time and spectral characteristics required in FSRS a series of linear and non-linear pulse conversion techniques were used. When compared to other FSRS systems reported so far, the constructed setup is very versatile: no-gap tuning of pulses from near ultraviolet to near infrared, high temporal (down to 40 fs) and spectral (down to 5 cm^{-1}) resolution, high output pulse energy (> 1 μJ), low noise (<4 μOD) and an expanded region of spectral components of the probe pulses (340-1030 nm). A new method for narrowing the spectrum of femtosecond pulses based on parametric amplification of chirped light was demonstrated that simplifies the setup and increases its efficiency .

Several issues, not previously discussed in context of FSRS technique, are discussed in the thesis including an internal calibration of spectra and a careful examination of Raman line profiles as a function of the delay between the probe and the Raman pulse. In the latter case, it was observed that change of the position of the narrow Raman line can be used to characterize the structure of the picosecond Raman pulse.

Vibrational transient states have been studied in three chemical (β-carotene, perylene, porphycene) and two biochemical systems (enhanced green fluorescent protein EGFP and the reversible photoswitching fluorescent protein Padron). High sensitivity of the transient Raman signal to experimental parameters was found for β-carotene. In perylene, a set of complete resonance Raman spectra S1 were acquired using several distinct wavelengths of the Raman pulse placed around transient absorption band at 700 nm. Also, modulations of the central position and amplitude of the bands with the frequencies of other mods were observed. The vibrational frequencies of S1 state in porphycene were obtained based on impulsive technique where the chirped white light pulse was used as a probe. In the case of EGFP, the characteristic modes of long-lived excited anionic form of a chromophore were determined - no significant evolution of the spectra was observed within the available time window. It was confirmed that the main process after excitation of the dark form of the Padron protein is the internal conversion within 4 ps and, in case of excitation of the bright form, transition to the fluorescent anionic cis form of the chromophore within 24 ps. A new component of about 200 fs in both forms of the Padron protein was discovered with characteristic activation of low frequency modes not observed in the ground electronic state.