Variable Magnetic-Field (VMF) Ultrafast Polarization Phase Selective (PPS) Attomechanics Studies in the Areas of Fundamental Significance to Biochemistry/Biophysics: New Methods and Tools for Monitoring Electronic Processes in Biosystems

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Introduction

This proposal to study electron-spin structures and dynamics in soft-matter compounds takes advantage of unique NHMFL capabilities, specifically the newly developed instrumentation combining short-pulse laser sources (Vitara) with high magnetic fields (25 T Split-helix magnet) in cell 5. During 2018 at LSU and during May and June 2018 at NHMF we tested new 400nm centered CW and UF VMF PPS configurations and new MF–PPS orientations for monitoring electronic processes in Biosystems, including nitrogenase enzymes. All these studies have never been done before.

Experimental

The developed instrumental configurations in cell 5 at NHMFL using Vitara pulses spectrally or harmonically separated, so that for example broadband ~400nm pulses are delivered to polarizers, quarter wave plates etc. Two or more PPS beams are generated in the same path configuration and directed into the observed sample in magnetic field. We use different types of detectors: CCD cameras, balanced detectors, and some PMTs. In addition reference beams are measured before and after sample. We compared the precision of spectral phase interference ("fringes") and direct difference measurements. In addition, high ratio polarizers are positioned at different points to clearly identify various polarization contributions. In December 2018, we have resolved the issue of sending beam up through the magnet: we successfully constructed a new tube, sample holder and alignment add-ons so that we can choose different directions of magnetic field, including the one with beam that goes upwards through magnet, parallel to the field. We completed and published the first study of the second P-cluster synthesis and the first study of the step one turnover (TO) in nitrogenase.

Results and Discussion

The last two experiments were also the beginning of the new project: we started measurements with new configuration for fs dynamics studies and tested responses to a variety of magnetic field orientations and strengths. We learned that so far our same path configuration provides most reliable phase-sensitive detection. We are investigating how precise is detection of *difference* between different PPS induced transitions in differently oriented magnetic fields using (1) spectral phase interference methods and (2) direct intensity difference measurements. Electron-spin dynamics studies in biomolecules can now include shorter fs time dynamics measurements and more detailed measurements of dependences on PPS-tailored electric and high magnetic fields. *In situ* measurements of biomolecular electron-spin dynamics will require high magnetic fields. New important scientific questions could be answered from our investigations of protein synthesis dynamics and TO steps in enzymes. Our novel PPS and EPR spectroscopic study of the biosynthesis of the "second" nitrogenase P-cluster[1] provides inside in a number of newly observed steps during that process as well as points to not yet completely resolved interactions that make these two P-clusters synthesis paths influence each other at a relatively large distance. Our new PPS measurements have also opened the view on the electron-spin structure of the first TO state of FeMoCo in nitrogenase which, as we found, has a paramagnetic contributions.

Conclusions

Developed methods will help clarify proposed electron-spin structures assignments and provide new dynamics models. For example, contrary to what have been suggested assumptions in a recently published study[2], results of our magnetic PPS spectra and magnetization study provide evidence that the ground state of the first TO state of FeMoCo nitrogenase[3] has paramagnetic contributions, including S=2. Such findings in combination with spin dynamics can redefine the previously proposed role of this and other Fe-S clusters in enzymes. Indeed, much better insights will come from our ongoing and future ultrafast dynamics studies. The most important questions now are: (1) what else can these PPS-magnetic methods see that is not accessible by other methods, particularly at the electron-spin spatiotemporal scales and (2) how to develop models that describe physical aspects of these attomehanics-level (controlled) events.

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References

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