

Jim Werner

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EDUCATION	Cornell University (Ithaca, NY)
1998	Ph.D. in Applied Physics
1994	M.S. in Applied Physics
1992	California Institute of Technology (Pasadena, CA)
	BS in Applied Physics

RESEARCH INTERESTS

Biophysics (cellular signaling processes, membrane dynamics, protein folding and conformational fluctuations) instrument development (microscopes, flow cytometry systems, microfluidics), laser spectroscopy (fluorescence, time-resolved, and Raman), and analytic and biophysical applications of single molecule detection by laser induced fluorescence.

WORK EXPERIENCE

Los Alamos National Laboratory, Technical Staff Member (2002-present)
Los Alamos National Laboratory, Postdoctoral Research Associate (1998-2002)
Cornell University, Hertz Foundation Fellow (1992-1998)

SKILLS

- **LASER SPECTROSCOPY:** Single molecule fluorescence spectroscopy, time correlated single photon counting, fluorescence correlation spectroscopy, four-wave mixing and nonlinear optics, optical trapping and manipulation, CW and pulsed laser sources from the vacuum ultra-violet to the mid IR.
- **OPTICAL MICROSCOPY:** Confocal and wide field fluorescence microscopy, including the full instrument development of a new, proprietary confocal microscope which employs a unique spatial filter geometry and active feed-back to follow individual fluorescent molecules in 3 spatial dimensions.
- **COMPUTER PROGRAMMING:** C/C++, Labview, Fortran, and Igor Pro. Instrument control using real-time operating systems. Experience with the writing and use of simple Monte Carlo simulations for data interpretation and instrument design.
- **PROGRAM DEVELOPMENT:** Have grown internal, highly competitive (~8% pay-line) proposal based funding into award-winning programs now supported through external sponsors, such as the National Institutes of Health. Submit, organize, and participate in a number of multi-investigator research proposal efforts each year.
- **SUPERVISORY EXPERIENCE:** Have mentored 5 postdoctoral research associates and assisted with the supervision of a number of summer research students.

SELECTED AWARDS

R&D 100 Award “3D Tracking Microscope” (2008)
Hertz Foundation Fellowship (1992-1997)
Caltech Carnation Merit Award (1990-1992)

CURRENT FUNDING

Title: *Tracking the Intracellular Spatial Trajectory of an Individual IgE Molecule*

Source: National Institutes of Health (NIH), R21AI07707

Role: PI

Duration: 1/2008 to 1/2010

Work follows IgE-FcεR1 receptor signaling, transport, and down-regulation using novel 3D single molecule tracking technology developed in our laboratory.

Title: *Predictive Design of Noble Metal Nanoclusters*

Source: Los Alamos Directed Research and Development-Directed Research (LDRD-DR)

Role: Co-investigator, PI: Jen Martinez

Duration: 10/2008 to 10/2011

Work aims to develop fluorescent noble metal nanoclusters for applications in biological imaging and sensing. My role involves single-molecule photophysical characterization of the clusters and testing their merit for use in cellular microscopy.

Title: *Center for Integrated Nanotechnologies*

Source: Department of Energy Basic Energy Sciences (DOE-BES)

Role: co-investigator, PI: Bob Hwang, Sandia National Laboratories

Duration: (on going, renewable funding)

My role in this nanotechnology center involves helping external users acquire and interpret data using conventional and home-built fluorescence microscopes, with some effort set aside for new technique development.

Title: *Understanding the Mechanism of Plant Virus-Vector Interaction through the use of New Imaging Technologies.*

Source: Office of Research, UC Riverside, UCR-LANL Collaborative program

Role: co-PI (other co-PI, James Ng, UC Riverside)

Duration: 10/1/2008 to 10/1/2009

This is seed-funding to foster collaborations in plant pathogen research between UC Riverside and Los Alamos National Laboratories and aims to better understand plant pathogen localization inside insect vector hosts.

Title: *Incorporating Simultaneous Wide-field Imaging with Confocal 3D Tracking*

Source: Los Alamos National Laboratory Technology Maturation

Role: PI

Duration: 10/1/2009 to 12/1/2009

This funding supports moving our 3D tracking microscope onto a more conventional inverted microscope platform, which should assist with commercialization efforts.

COMPLETED PROJECTS

Title: *Photophysical Characterization of Probes for 3D Molecular Tracking*

Source: National Nanotechnology Enterprise Development Center (NNEDC).

Role: PI

Duration: 10/2005 to 10/2007

Work tested various fluorescent reporters (quantum dots, organic dyes, fluorescent metal nanoclusters) for use in 3D intracellular tracking applications. Work resulted in continued development for our 3D tracking efforts and helped bridge the time-gap between the LDRD and NIH funding. NNEDC funds are not eligible for renewal.

COMPLETED PROJECTS (Continued)

Title: *Measuring Individual Antigen-Antibody Encounters for Decades of Timescales*

Source: Los Alamos Directed Research and Development- Exploratory Research (LDRD-ER)

Role: PI

Duration: 10/2004 to 10/2007

Work developed methods (surface functionalization and image analysis software) needed to observe individual antigen binding events to surface immobilized antibodies above the background of non-specific adsorption to the surface. Work led to a number of publications and provided the preliminary data needed for competitive proposal submissions to the NIH and the Defense Threat Reduction Agency (DTRA). LDRD funds are not eligible for renewal.

Title: *A Microfabricated, Single Molecule Sorter*

Source: Los Alamos Directed Research and Development- Exploratory Research (LDRD-ER)

Role: PI

Duration: 10/2004 to 10/2007

Work tested various flow chamber geometries for molecular sorting applications. This project helped support an experimental demonstration that single molecule energy transfer distributions measured in flow can be of superior resolution than distributions measured via diffusion through an open confocal volume element. LDRD funds are not eligible for renewal.

Title: *Can Single Molecules be Tracked in Cells?*

Source: Los Alamos National Laboratory Technology Maturation Funds

Role: PI

Duration: 10/2005 to 10/2006

Work determined the background levels, speed, and tracking accuracy of our 3D tracking methods, with special attention to whether our tracking methods could work in a “dirty” cellular environment. Technology Maturation Funds are not eligible for renewal.

Title: *Tracking Single Molecules in Three Dimensions*

Source: Los Alamos Directed Research and Development- Exploratory Research (LDRD-ER)

Role: PI

Duration: 10/2002 to 10/2005

Work developed instrumentation (a confocal microscope that uses active-feedback and a unique spatial filter geometry) needed to track individual molecules through 3 spatial dimensions. Demonstrated tracking of individual quantum dots undergoing Brownian diffusion at rates faster than most cellular signaling processes. Work led to publications, patents, and funding from the NIH, NNEDC, and Technology Maturation funds. LDRD funds are not eligible for renewal.

Title: *Demonstration of Quantum Entanglement in the Solid State*

Source: Los Alamos Directed Research and Development- Directed Research (LDRD-DR)

Role: co-investigator, PI: Marilyn Hawley

Duration: 10/2003 to 10/2006

Work tested the feasibility of optical readout to determine the spin state of individual phosphorous atoms in a silicon matrix, which has applications in quantum computation.

COMPLETED PROJECTS (Continued)

Title: *Biosensors Based Upon Biomimetic Recognition*

Source: Los Alamos Directed Research and Development- Directed Research (LDRD-DR)

Role: co-investigator, PI: Basil Swanson

Duration: 10/2002 to 10/2005

Work explored biomolecular recognition based upon proximity-based co-localization and energy transfer of recognition elements imbedded in a planar lipid bilayer. My work led to a patent and two different competitive proposal submissions to the NIH. LDRD funds are not eligible for renewal.

Title: *Single Molecule Protein Folding*

Source: Los Alamos Directed Research and Development- Directed Research (LDRD-DR)

Role: co-investigator, PI: Richard Keller

Duration: 10/2002 to 10/2005

Work developed methods for examining individual protein conformation and folding dynamics. This project led to a number of peer-reviewed publications and provided the preliminary data for proposal submissions to the NIH. LDRD funds are not eligible for renewal.

SERVICE

- Reviewer for *Nature Methods*, *Biophysical Journal*, *Analytical Chemistry*, *Journal of Physical Chemistry*, *Applied Optics*, and *Cytometry*.
- Served on Laboratory Directed Research and Development (LDRD) proposal review panels in Biology, Biochemistry & Biophysics (BBB) for FY2005, as well as Atomic and Molecular Optical Physics (AMOP) for FY2004.
- Chaired a session on *Biological and Non-Biological Optical Microscopy* for the National OSA meeting (Frontiers in Optics XIX) San Jose, CA, September 20, 2007.

SELECTED ORAL PRESENTATIONS

"Tracking Single Quantum Dots in 3 Dimensions," **Frontiers in Optics: Laser Science XIX**, San Jose, CA, September 20, 2007. (Invited)

"Three dimensional tracking of single quantum dots," **Nanotechnology in Cellular Biology**, University of New Mexico IGERT symposium, Albuquerque, NM, August 14, 2007. (Invited)

"Three dimensional tracking of individual quantum dots," **27th Annual Meeting of the Center for Nonlinear Studies**, Santa Fe, NM, May 2007. (Invited)

"Tracking single quantum dots in 3 dimensions" **Aspen Center for Physics, Single Molecule Biophysics**, Aspen, CO, February 2007.

"An approach to tracking single fluorophores in 3-D" **Portland State University Chemistry Department Seminar Series**, Portland, OR, October 2005. (Invited)

"An approach to tracking single molecules in 3-D" delivered at **Pittcon**, Orlando, FL, March 2005. (Invited)

"Single molecule tracking," **Sandia National Laboratories Seminar Series**, Albuquerque, NM, August 2004. (Invited)

"Energy transfer distributions measured by single molecule fluorescence flow cytometry," **Aspen Center for Physics, Single Molecule Biophysics**, Aspen, CO, January 2003

"Dynamics of the primary processes in protein folding: Helix nucleation." **American Chemical Society Annual Meeting**, San Diego, CA, April 2002. (Invited)

"Current status of DNA sequencing by single molecule detection," **Photonics West**, San Jose, CA, January 1999.

PUBLICATIONS

- [32] Werner, J. H., Montano G.A., Garcia A.L., Zurek N.A., Akhadov E.A., Lopez G.P. and Shreve A.P., "Formation and dynamics of supported phospholipid membranes on a periodic nanotextured substrate," **Langmuir** (2009, in press).
- [31] Wells, N. P., Lessard G.A., Phipps M.E., Goodwin P. M., Lidke D.S., Wilson B.S. and Werner J. H., "Going Beyond 2D: Following membrane diffusion and topography in the IgE-Fc[Epsilon]RI system using 3-dimensional tracking microscopy," **Proc. of the SPIE** 7185-1 to 7185-13 (2009, in press).
- [30] Wells, N. P., Lessard G.A. and Werner J. H., "Confocal, 3-Dimensional Tracking of Individual Quantum-Dots in High Background Environments," **Analytical Chemistry** 9830-9834 (2008).
- [29] Temirov, J., Bradbury A. and Werner J. H., "Measuring an Antibody Affinity Distribution Molecule by Molecule," **Analytical Chemistry** 8642-8648 (2008).
- [28] Lessard, G.A., Habuchi S, Werner J.H., Goodwin P. M., De Schryver F.C., Hofkens J. and Cotlet M., "Probing dimerization and Intraprotein fluorescence resonance energy transfer in a far red fluorescent protein from the sea anemone *Heteractis crispa*," **Journal of Biomedical Optics** 031212-1 to 031212-7 (2008).
- [27] Dai, M., Temirov J., Emanuele P., Kiss C., Pavlik P., Werner J. H. and Bradbury A., "Using T7 phage display to select GFP-based binders," **Protein Engineering Design and Selection** (2008).
- [26] Lessard, G.A., Goodwin P. M. and Werner J. H., "Three dimensional tracking of individual quantum dots," **Applied Physics Letters** 2224106 (2007).
- [25] Werner, J. H., Mccarney E. R., Keller R. A., Plaxco K. W. and Goodwin P. M., "Increasing the resolution of single pair fluorescence resonance energy transfer measurements in solution via molecular cytometry," **Analytical Chemistry** 3509-3513 (2007).
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- [23] Werner, J. H., Joggerst R, Dyer R.B. and Goodwin P. M., "A two dimensional view of the folding energy landscape of cytochrome c," **Proceedings of the National Academy of Sciences (USA)** 11130-11135 (2006).
- [22] Temirov, J, Bradbury A and Werner J.H., "Surface-immobilized antibody-antigen binding affinity studies by single molecule fluorescence imaging," **Proceedings of the SPIE** 6092001 to 60920010 (2006).
- [21] Lessard, G. , Goodwin P.M. and Werner J.H., "Three dimensional tracking of single fluorescent particles," **Proceedings of the SPIE** 609205-1 to 609205-8 (2006).
- [20] Cotlet, M. , Habuchi S., Whitier J. E., Werner J. H., De Schryver F. C., Hofkens J. and Goodwin P. M., "Single Molecule Spectroscopic Characterization of a Far-Red Fluorescent protein (HcRed) from the Anthozoa Coral *Heteractis Crispa*," **Proc. of the SPIE** 609804-1 to 690804-11 (2006).
- [19] Cotlet, M., Goodwin P.M., Waldo G. S. and Werner J. H., "Time-resolved Detection of the One- and Two-Photon Excited Fluorescence of Single Molecules of a Folding Enhanced Green Fluorescent Protein," **Proc. of the SPIE** 609204-1 to 609204-10 (2006).
- [18] Cotlet, M., Goodwin P.M., Waldo G. S. and Werner J. H., "A comparison of the fluorescence dynamics of single molecules of a green fluorescent protein: one- vs two-photon excitation," **ChemPhysChem** 250-260 (2006).

- [17] Werner, J.H., Cai H., Keller R.A. and Goodwin P.M., "Exonuclease I hydrolyzes DNA with a distribution of rates," **Biophysical Journal** 1403-1412 (2005).
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- [15] Werner, J.H., Cai H., Jett J.H., Reha-Krantz L., Keller R.A. and Goodwin P.M., "Progress towards single-molecule DNA sequencing: A one color demonstration," **Journal of Biotechnology** 1-14 (2003).
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- [13] Baker, G.A., Baker S.N., Mccleskey T.M. and Werner J.H., "Aspects of chemical recognition and biosolvation within room temperature ionic liquids," **ACS Symposium Series: Ionic Liquids as Green Solvents** 212-224 (2003).
- [12] Werner, J.H., Baker S.N. and Baker G.A., "Fluorescence correlation spectroscopic studies of diffusion within the ionic liquid 1-butyl-3-methylimidazolium hexafluorophosphate," **Analyst** 786-789 (2003).
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- [10] Goodwin, P.M., Ambrose W.P., Cai H., Grace W.K., Larson E.J., Marrone B.L., Jett J.H., Werner J.H. and Keller R. A., "Single molecule nucleic acid analysis by fluorescence flow cytometry," **NATO Advanced Research Workshop on Biological, Biophysical and Theoretical Aspects of Polymer Structure and Transport** 351-370 (2002).
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- [3] Werner, J.H. and Cool T.A., "Flame sampling photoionization mass spectrometry of dichloroethanol.," **Chemical Physics Letters** 81-7 (1998).
- [2] Werner, J.H. and Cool T.A., "Flame sampling photoionization mass spectrometry of CH₃PO₂ and CH₃OPO₂.,," **Chemical Physics Letters** 278-82 (1997).
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PATENTS & INVENTION DISCLOSURES

Werner, J. H., Shreve A.P. and Goodwin P. M., "*Method and Apparatus for 3D imaging on nanometer length scales*," **LANL Invention Disclosure S112966/L2008060** (2008).

Werner, J.H., Goodwin P. M. and Lessard G., "*Apparatus and method for tracking a molecule or particle in three dimensions*," **US Patent Application 20080085550** (2008).

Werner, J.H., Temirov J and Bradbury A., "*Single-molecule antibody arrays*," **LANL invention disclosure S-109,108** (Filed internally 6/2005).

Werner, J. H., Reed S.M. and Swanson B. I., "*Screening of libraries using fluorescently reactive lipids and fluorescence correlation spectroscopy*," **US Patent Application 20050191705** (Filed internally 4/2003.).

Cai, H., Goodwin P. M., Keller R. A. and Werner J. H., "*Rapid Haploid Genotyping by Single Molecule Detection*," **US Patent Application 20060008799** (Filed internally 5/2001.).