

# KEVIN C. CROSBY

*Department of Pharmacology  
University of Colorado School of Medicine,  
Anschutz Medical Campus, RC-1 North, P18-6402K  
12800 East 19<sup>th</sup> Ave., Aurora, CO 80045, United States  
E-mail: kevin.crosby@cuanschutz.edu*

## SUMMARY

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Multi-disciplinary expertise in the technical aspects of a range of advanced quantitative fluorescence microscopy methods, computational image analysis, and biophysics and molecular cell biology.

Specific experience in the use and analysis of multiple modes of fluorescence microscopy, such as super-resolution microscopy (PALM/STORM, STED, SIM), fluctuation-based techniques (FCS, SpIDA), FRET (both lifetime and intensity based), and quantitative FRAP. Proficiency in MATLAB and scripting for ImageJ/FIJI .

Working knowledge of a range of biological systems. Led projects in the realms of nano-scale organization of synaptic structure and signaling domains in excitatory neurons, the biophysics of membrane-associated proteins in mammalian cells, and plant cell biology centered on the *in vivo* analysis of localization, interactions, and complex formation of biosynthetic enzymes.

## Current Research Position

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### DEPARTMENT OF PHARMACOLOGY, UNIVERSITY OF COLORADO MEDICAL SCHOOL

*Research Instructor: 2022 – Present  
Research Associate: 2018 – 2022*

## RESEARCH EXPERIENCE

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### MARK L. DELL'ACQUA GROUP, DEPARTMENT OF PHARMACOLOGY, UNIVERSITY OF COLORADO MEDICAL SCHOOL

*Postdoctoral Fellow: July 2017 – 2018*

*National Institutes of Health NIAAA T32 funded Postdoctoral Fellow, 2014 – 2017*

**Research:** Organization, regulation, and dynamics of nano-signaling domains in the post-synaptic spine; biophysical mechanisms of synapse-to-nucleus communication; application of advanced fluorescence microscopy techniques to the study of neuronal cell biology.

**Techniques:** Super-resolution microscopy (PALM/STORM, STED, SIM), FCS, SpIDA, FLIM-FRET, quantitative FRAP Advanced statistical analysis of single-molecule super-resolution data (static and dynamic). Computational analysis (MATLAB, FIJI, IgorPro, basic R and Python knowledge). Rodent hippocampal dissections, primary neuronal cell culturing techniques. Molecular cloning techniques.

### T.W.J. GADELLA GROUP, SECTION MOLECULAR CYTOLOGY, VAN LEEUWENHOEK CENTRE FOR ADVANCED MICROSCOPY, UNIVERSITY OF AMSTERDAM

*European Science Foundation/NWO funded Postdoctoral Fellow, 2009 – 2013*

**Research:** Biophysics of the Ca<sup>2+</sup> responsive phospholipid binding annexin proteins; Gαq transduced GPCR signaling. Optimization and application of advanced fluorescence microscopy techniques for the study of membrane biology.

**Techniques:** Super-resolution photoactivated localization microscopy (PALM); fluorescence fluctuation based techniques, including fluorescence (cross) correlation spectroscopy (FCS) and number and brightness analysis (N&B); quantitative fluorescence recovery after photobleaching (FRAP); fluorescence lifetime imaging microscopy (FLIM), both frequency-domain and time-domain; total internal reflection microscopy (TIRF); Förster resonance energy transfer (FRET); mammalian cell culture and transfection.

*National Science Foundation (NSF) IGERT sponsored visiting Ph.D. Student, 2008*

**Research:** Use of fluorescence lifetime imaging microscopy FLIM-FRET to study interactions among flavonoid enzymes in *Arabidopsis* protoplasts.

## **B.S.J. WINKEL GROUP, DEPARTMENT OF BIOLOGICAL SCIENCES, VIRGINIA TECH**

*NSF IGERT pre-doctoral Fellow/Researcher, 2005 – 2009*

**Research:** *In vivo* analysis of localization, interactions, and complex formation of the *Arabidopsis* flavonoid biosynthetic enzymes. *In planta* evaluation of *Arabidopsis* flavonol synthase gene function.

**Techniques:** Confocal live cell imaging and immunofluorescent microscopy; Protoplast isolation and transfection; HPLC and spectrophotometric analysis of metabolites; Standard molecular biology and biochemical methods.

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## EDUCATION

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### **VIRGINIA POLYTECHNIC INSTITUTE AND STATE UNIVERSITY**

*Ph.D. Department of Biological Sciences, December 2008*

**Thesis:** Organization and function of subcellular macromolecular complexes: A multi-system analysis

**Advisor:** Professor Brenda S.J. Winkel

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## GRANTS AND FELLOWSHIPS

### **INTERNATIONAL VISITING SCIENTIST AWARD**

*National Institutes of Health (Broadening Experiences in Scientific Training program). Funds to support collaborative visit to **Professor Paul Wiseman**'s group at **McGill University, Montreal** to apply techniques in Image Correlation Spectroscopy and Spatial Intensity Distribution Analysis to study assembly of post-synaptic regulatory complexes. September 2016 – October 2016*

### **POSTDOCTORAL TRAINING FELLOWSHIP**

*National Institutes of Health (T32), NIAAA Institutional Research Training Program*

*July 2014 – July 2017*

### **PH.D. TRAINING FELLOWSHIP**

*The EIGER (Exploring Interfaces Through Graduate Education and Research) Project, U.S. National Science Foundation, Integrative Graduate Education and Research Traineeship (IGERT) program.*

*December 2005 – December 2008*

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## TEACHING AND ADVISING EXPERIENCE

### **THE CAJAL ADVANCED NEUROSCIENCE TRAINING PROGRAMME: ADVANCED TECHNIQUES FOR SYNAPTIC BIOLOGY; BORDEAUX SCHOOL OF NEUROSCIENCE**

*23 October – 10 November 2023*

*Project Instructor:* Provided instruction and guidance during a ten-day project focusing on multiple single-molecule labeling and imaging techniques.

### **DEPARTMENT OF CLINICAL AND EXPERIMENTAL MEDICINE, IKE, LINKÖPING UNIVERSITY, SWEDEN**

*Invited Lecturer:* Advanced Light Microscopy and Live Cell Imaging Course. April 2013.

### **VAN LEEUWENHOEK CENTRE FOR ADVANCED MICROSCOPY, SWAMMERDAM INSTITUTE FOR LIFE SCIENCES, UNIVERSITY OF AMSTERDAM**

*Project supervisor:* Two Masters Students with nine month rotations and one advanced Bachelors student in the Erasmus Program – six month internship. All three went on to pursue a Ph.D.

*Instructor:* International and Masters track microscopy courses. Practical instruction on techniques such as super-resolution (photoactivated localization microscopy - PALM), total internal reflection microscopy (TIRF), and fluorescence lifetime imaging microscopy (FLIM).

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## PUBLICATIONS

(†First author or co-contributing primary author):

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Samantha S. Olah, Dean J. Kareemo, William C. Buchta, Brooke L. Sinnen, Carley N. Miller, Hannah S. Actor-Engel, Sara S. Gookin, Christina S. Winborn, Mason S. Kleinjan, Kevin C. Crosby, Jason Aoto, Katharine R. Smith, Matthew J. Kennedy. **Acute reorganization of postsynaptic neurotransmitter receptors reveals the functional impact of molecular nanoarchitecture at GABAergic inhibitory synapses.** *Cell Reports*, (in press).

Harrison J Ramsay, Sara E Gookin, Austin M Ramsey, Dean J Kareemo, Kevin C Crosby, Dominik G Stich, Samantha S Olah, Hannah S Actor-Engel, Katharine R Smith, Matthew J Kennedy. **AMPA and GABAA receptor nanodomains assemble in the absence of synaptic neurotransmitter release.** *Front. Mol. Neurosci.*, 2023, 16.

Xiaobing Chen, Kevin C. Crosby, Austin Feng, Alicia M. Purkey, Maria A. Aronova, Christine A. Winters, Virginia T. Crocker, Richard D. Leapman, Thomas S. Reese, and Mark L. Dell'Acqua. **Palmitoylation of A-kinase anchoring protein 79/150 modulates its nanoscale organization, trafficking, and mobility in postsynaptic spines.** *Front. Synaptic Neurosci.*, 2022, 15.

Sara E. Gookin, Matthew R. Taylor, Samantha L. Schwartz, Matthew J. Kennedy, Mark L. Dell'Acqua, Kevin C. Crosby, and Katharine R. Smith. **Complementary use of super-resolution imaging modalities to study the nanoscale architecture of inhibitory synapses.** *Frontiers in Synaptic Neuroscience*, 2022, 14: 852227.

Joshua D. Garcia, Sara E. Gookin, Kevin C. Crosby, Samantha L. Schwartz, Erika Tiemeier, Matthew J. Kennedy, Mark L. Dell'Acqua, Paco S. Herson, Nidia Quillinan, and Katharine R. Smith. **Stepwise disassembly of GABAergic synapses during pathogenic excitotoxicity.** *Cell Reports*, 2021, 37(12) 110142.

Hannah S. Actor-Engel, Samantha L. Schwartz, Kevin C. Crosby, Brooke L. Sinnen, Olga Prikhodko, Harrison J. Ramsay, Jennifer N. Bourne, Christina S. Winborn, Alexandra Lucas, Katharine R. Smith, Mark L. Dell'Acqua, and Matthew J. Kennedy. **Precision mapping of Amyloid-Beta binding reveals perisynaptic localization and spatially restricted plasticity deficits.** *Eneuro*, 2021, 8(6).

Carolyn Nicole Brown, Sarah G. Cook, Hillary F. Allen, Kevin C. Crosby, Tarjinder Singh, Steven J. Coultrap, and K. Ulrich Bayer. **Characterization of six CamKII $\alpha$  variants found in patients with schizophrenia.** *iScience*, 2021, 24(10) 103184.

† Jonathan G. Murphy\*, Kevin C. Crosby\*, Phillip J. Dittmer, William A. Sather, and Mark L. Dell'Acqua. **AKAP79/150 recruits the transcription factor NFAT to regulate signaling to the nucleus by neuronal L-type Ca<sup>2+</sup> channels.** *Molecular Biology of the Cell*, 2019, 30(14) 1743-1756; (\*KCC and JGM contributed equally to this manuscript).

† Kevin C. Crosby, Sara E. Gookin, Joshua D. Garcia, Katlin M. Hahm, Mark L. Dell'Acqua, and Katharine R. Smith. **Nanoscale subsynaptic domains underlie the organization of the inhibitory synapse.** *Cell Reports*, 2019, 26(12) 3284-3297.

Alicia M. Purkey, Kevin M. Woolfrey, Kevin C. Crosby, Dominik G. Stich, Wallace S. Chick, Jason Aoto, and Mark L. Dell'Acqua. **AKAP150 Palmitoylation regulates synaptic incorporation of Ca<sup>2+</sup>-permeable AMPA receptors to control LTP.** *Cell Reports*, 2018, 25(4) 974-987.

Jonathan O. Watkinson, Peter A. Bowerman, Kevin C. Crosby, Sherry B. Hildreth, Richard F. Helm, and Brenda S.J. Winkel. **Identification of MOS9 as an interaction partner for chalcone synthase in the nucleus.** *PeerJ*, 6 e5598.

Brooke L. Sinnen\*, Aaron Bowen\*, Jeffery S. Forte\*, Brian Heister\*, Kevin C. Crosby, Emily Gibson, Mark L. Dell'Acqua, and Matthew J. Kennedy. **Optogenetic control of synaptic composition and function.** *Neuron*, 2016, 93(3): 646-660. (\*contributed equally to this manuscript).

† Eelco Hoogendoorn\*, Kevin C. Crosby\*, Daniela Leyton-Puig\*, Ronald M.P. Breedijk, Kees Jalink, Theodorus W.J. Gadella, Jr., and Marten Postma. **The fidelity of stochastic single-molecule super-resolution reconstructions critically depends upon robust background estimation.** *Sci Rep*, 2014,

4(3854) doi: 10.1038/srep03854. (\*EH, KCC, and DLP contributed equally to this manuscript).  
\*Selected as a Research Highlight in: Daniel Evanko, **Taming the image background beast.** *Nat Meth*, 2014, 11(3): 228.

† Kevin C. Crosby, Marten Postma, Mark A. Hink, Christiaan H.C. Zeelenberg, Merel J.W. Adjobo-Hermans, and Theodorus W.J. Gadella, Jr. **Quantitative analysis of self-association and mobility of annexin A4 at the plasma membrane.** *Biophys J*, 2013, 104(9): 1875-1855. \* Highlighted as New and Notable in: Andrew H.A. Clayton and Amitabha Chattopadhyay. **A toolbox of fluorescence microscopy approaches reveals dynamics and assembly of a membrane-associated protein.** *Biophys J* 2013, 104(9): 1844-1845.

Merel J.W. Adjobo-Hermans, Kevin C. Crosby, Mateusz Putyrski, Arshia Bhageloe, Laura van Weeren, Carsten Schultz, Joachim Goedhart, and Theodorus W.J. Gadella Jr. **PLC $\beta$  isoforms differ in their subcellular location and their CT-domain dependent interaction with G $\alpha$ q.** *Cell Signal* 2013, 25(1): 255-63.

† Kevin C. Crosby, Anna Pietraszewska-Bogiel, Theodorus W.J. Gadella, Jr., and Brenda S.J. Winkel. **Förster resonance energy transfer demonstrates a flavonoid metabolon in living plant cells that displays competitive interactions between enzymes.** *FEBS Lett* 2011, 585(14): 2193-8.

† Daniel K. Owens\*, Anne B. Alerding\*, Kevin C. Crosby\*, Aloka B. Bandara, James H. Westwood, and Brenda S.J. Winkel. **Functional analysis of the *Arabidopsis* flavonol synthase gene family.** *Plant Physiology* 2008, 143(3); 1046-61. (\*DKO, ABA, and KCC contributed equally to this manuscript).

Daniel K. Owens, Kevin C. Crosby, Justin Runac, Brad A. Howard, and Brenda S.J. Winkel. **Biochemical and genetic characterization of *Arabidopsis* flavanone 3 $\beta$ -hydroxylase.** *Plant Physiol. and Biochem* 2008, 46(10); 833-43.