BIOGRAPHICAL SKETCH

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Follow this format for each person. **DO NOT EXCEED FIVE PAGES.**

NAME: Marriah Green

eRA COMMONS USER NAME (credential, e.g., agency login): MARRIAHGREEN

POSITION TITLE: Graduate Student Research Assistant

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Start Date MM/YYYY	Completion Date MM/YYYY	FIELD OF STUDY
Palomar College 1140 West Mission Road San Marcos, CA 92069	NA	06/2010	01/2011	NA
MiraCosta College 1 Barnard Drive Oceanside, CA 92056	Associate in Arts (A.A)	08/2011	06/2014	A.A. in Biological Sciences A.A. in Liberal Arts Emphasis in Mathematics and Sciences
University of California San Diego 9500 Gilman Dr La Jolla, CA 92093	BS	09/2014	01/2017	Biochemistry/ Chemistry
Columbia University, Graduate School of Arts and Sciences 535 W 116th St #109 New York, NY 10027	MA MP PhD	08/2017 08/2017 08/2017	02/2019 05/2020 Present	Nutritional and Metabolic Biology Nutritional and Metabolic Biology

A. Personal Statement

My long-term research and career interests lie in elucidating the structures and functions of integral membrane proteins related to human health and disease as an independent academic researcher. I seek to utilize single-particle cryogenic electron microscopy (Cryo-EM) and X-ray crystallography to determine the structure of biomedically relevant proteins, specifically ion channels to provide novel functional insights into neurotransmission. My academic training and research experience to date have provided me with a broad background in chemistry, molecular biology, biochemistry, and structural biology. As an undergraduate, I had to work multiple jobs to put myself through college, yet I still managed to gain research experience. While studying biochemistry in the chemistry department at University of California, San Diego (UCSD), I conducted research at The Scripps Research Institute (TSRI) and at Cornell University. These initial scientific experiences introduced me to laboratory research and exposed me to different areas of research. This allowed me the opportunity to not only explore related fields but more importantly, solidified my desire to become an academic researcher.

At UCSD, there were some personal issues that had adverse effects on my grades. While I did not want to stop my education, these issues did affect my grades in 2017. I recognize that my grades may appear to be a weakness; regardless of my learning disabilities, I have illustrated my capability of improved grades due to my strength and resilience. My imperfect grades are a testament of dedication to my education. Despite my disadvantages, I have persevered, working my way through college in addition to heavy course work and research internships. My passion for scientific discovery remains persistent and I continue to be tenaciously

driven in working towards my scientific career aspirations. Due to one of my learning disabilities (I have been officially diagnosed with dyslexia), some weaknesses arise in my grades and writing. To proactively address this issue, I attended the Funding and Grantsmanship for Research and Career Development Activities Course instructed by Dr Jaime Rubin. I will continue to improve my writing and career development by working with my mentor, Dr. Alexander Sobolevsky, as well as Dr. Jaime Rubin. My ultimate career development goals are to become a principle investigator at an academic institution, where I would cultivate an environment that is accepting of all learning styles and encourage students with learning disabilities that they are not disabled from pursuing their dreams. As an academic researcher, I will continue to provide research opportunities to underrepresented students, encourage campus diversity by participating in admissions, and support programs such as Graduates Initiative for Diversity (GID), to influence the next generation of budding scientists from diverse backgrounds to achieve their academic and scientific career goals. Thus far, I have helped Columbia University Irving Medical Center (CUIMC) promote diversity by participating as a proactive board member with the GID organization.

For my doctoral training at CUIMC I joined the structural biology lab of Dr. Sobolevsky. In the Sobolevsky lab I carry out the proposed research on elucidating the structure and function of the ionotropic glutamate receptor (iGluR) homologue, glutamate like receptor (GLR). Understanding how these vital proteins function and relate to iGluRs is important for insight into glutamate receptors, iGluRs expressed in non-neuronal cells, and the evolution of calcium signaling. Calcium signaling is an example of fundamental signaling for all eukaryotic cells and abnormalities in signaling are involved in numerous pathologies from cancer to neurodegenerative diseases. Thus, studying GLRs as calcium permeable ion channels will increase our understanding of this complex biological process.

Investigating GLR structure and function will require the application of techniques in biochemistry, biophysics, electrophysiology, and structural biology, including X-ray crystallography as well as cryogenic electron microscopy (cryo-EM). Dr. Sobolevsky, a recognized leader in iGluR structure and function, uses all of these techniques to conduct cutting-edge research of ion channels and thus I am well positioned to accomplish my projected goals. The Sobolevsky lab is highly adept at membrane protein purification methods and functional characterization, since joining the lab I have developed skills in eukaryotic protein expression, optimization of protein purification, functional assays, and learned new structural biology techniques including X-ray crystallography and Cryo-EM. My research progress on the crystal structure of the GLR ligand binding domain has resulted in a co-first authorship. In progress is another first authorship for my study on the function and full length GLR structure. The Sobolevsky lab also collaborates with the New York Structural Biology Center (NYSBC), which is at the forefront of protein structure determination by cryo-EM. I have also been fortunate enough to attend NYSBC courses and discussion groups, and have also been trained in Cryo-EM related technology by the highly trained NYSBC affiliates. I am confident that my past research experiences as well as the excellent resources and expertise available to me at both CUIMC and NYSBC will allow me to successfully complete my proposed project and purse a career as an independent scientist.

I have the motivation and potential to carry out the proposed work as my academic coursework and training expertise have provided me with an excellent understanding of molecular biology, physiology, biochemistry, and a variety of structural biology techniques. If awarded, this fellowship would support the continuation of my proposed research project, choice of sponsor and training to enhance my potential for a productive independent scientific research career.

B. Positions and Honors

Positions

2012-2014 Learning Center Tutor, MiraCosta College, Highest Level of Tutor for Chemistry, Microbiology, Biology, Calculus, Organic Chemistry, and Algebra
2013-2014 Chemistry Teaching Assistant
2014 MiraCosta Science Fair
2015 Undergraduate Research Assistant, University of California, San Diego, Lab of Dr. Farquhar
2015 Cornell University Food Science Summer Scholar Program
2016 Research Intern, The Scripps Research Institute, Lab of Dr. Sharpless
2017- PhD Candidate, Columbia University
2019- CUIMC Woman in Science at Columbia (WISC) Board Member, Position: Communications Director and Marketing Chair
2019 Group Leader, Girls Science Day Volunteer, Columbia University
2020- CUIMC Graduate Initiative for Diversity (GID) Board Member, Position: Social Networking Chair

Other Experiences

2014-2015 American Chemical Society Student Affiliates (ACSSA) Member
2015 International Society for Pharmaceutical Engineering (ISPE)
2014-2016 Biochemistry Club University of California, San Diego
2015 Institute of Food Technologists (IFT) Conference
2017-2019 The 20th – 22nd Annual Institute of Human Nutrition Retreat and Wu Lectureship
2017- NMB Liaison Committee Member
2017- CUIMC Woman in Science at Columbia (WISC) Participant
2018 Ionotropic Glutamate Receptor (iGluR) Retreat
2018- New York Structural biology Discussion Group Participant
2019 Presented Thesis Research at Nutrition Departmental Seminar
2019- Girls Science Day Group Leader, Columbia University
2019- Knitting Instructor at CUIMC Student Wellness Center
2019- Mentor to Undergraduates
2021 Biophysical Society Conference and Poster Presentation
2021 WISC Conference and Poster Presentation

Honors and Awards

2011-2013 Honors Scholar Program, MiraCosta College
2011-2014 Permanent Honor Roll, Member of Phi Theta Kappa Honors Society, and President's List, MiraCosta College
3012-2013 MiraCosta Muriel Kaplan Scholarship
2013-2014 MiraCosta College Linda Koelkebech Memorial Merit Scholarship
2014 Highest Honors Certificate of Achievement in IGETC, MiraCosta College
2014-2015 Provost Honors, University of California, San Diego
2016 American Chemical Society Certification Award, University of California, San Diego
2017 Columbia Minority Supplement, Provost Diversity Fellowship
2017-2018 Graduate Training in Nutrition (Project number: 5T32DK007647-29)

C. Contributions to Science

C.1 Undergraduate Research at Cornell University My first fulfilling undergraduate research experience was in 2015, when I was awarded a position in the Food Science Summer Scholar Program at Cornell University. There I conducted research with Dr. Elad Tako studying micronutrient metabolism and physiology at the United States Department of Agriculture - Agricultural Research Service (USDA-ARS) Trace Minerals and Nutrition Unit. I found it meaningful to investigate iron deficiency and work towards discoveries to help people suffering from anemia in developing countries. At the end of the Program, I presented my project and findings at the final Food Science Department seminar open to the whole department. I submitted my abstract for this project and it was printed in the seminar's booklet, also open to all in the Department.

C.2 Undergraduate Research at The Scripps Research Institute In 2016 my research laboratory experience expanded from micronutrient metabolism to pharmacology during my last year of undergraduate when I landed a competitive research position. For half a year I participated in drug delivery and design research at Nobel Laureate Dr. K. Barry Sharpless's lab at The Scripps Research Institute (TSRI) in the Department of Chemistry. I worked to identify covalent drug candidates using a new aryl fluorosulfate small molecular library. To make a multifunctional molecule I took advantage of multivalent electrophilic molecular core, onto which an aryl fluorosulfate, an alkyne "pull-down", and a variable functional group were installed sequentially. Through synthetic chemistry, I helped build a small molecular library specifically with aryl fluorosulfate moieties incorporated. I assisted in the creation dendrimers for improving drug delivery, specifically increasing antitumor activity and chemotherapeutics' ability to pass the blood brain barrier. Future work on the project would include biological assays for screening these compounds to identify drug candidates.

C.3 Graduate Research at Columbia University I am currently conducting my thesis research in the lab of Dr. Alexander Sobolevsky at the Columbia University Irving Medical Center. I am using both structural and functional techniques to characterize the structure and function of the elusive Glutamate Receptor-Like (GLR) ion channels. Initially, I focused on a single domain of GLRs, the ligand binding domain (LBD), homologous to iGluRs' LBD which binds activating ligands and initiates gating in neuronal iGluRs. I was able to subclone various GLR LBDs into a plasmid for large-scale expression in bacterial cells and purify sufficient amounts of protein for crystallographic studies. Thus far, we succeeded in obtaining crystals of the GLR3.2 LBD in the presence of various ligands and solved a high-resolution structure using macromolecular X-ray crystallography. This work, including our crystal structures of the GLR LBD, has resulted in publication on which I am co-first author. We have also started structural studies of the full-length GLR using cryo-EM and successfully solved the first 3D

structure. We aim to solve the full-length GLR structure in different functional states at high resolution and to improve the resolution past 4 Å, our most current resolution. This study has led to our recent publication (a co-first author manuscript) on GLR function and structure revealing the first full-length structure. No full length GLR structure had previously been reported, so our results unveiled novel structural and functional information about this elusive protein. A GLR structure would lead to valuable insight to help understand how GLRs play a role in the cellular uptake of calcium, improving our understanding of plant neuronal like networks. Overall, studying GLR structure and function will provide basic research to enhance our understanding of ion channels, impact the field of calcium signaling at a fundamental level, offer functional insight of non-neuronal iGluRs and inform new strategies for treatment of human disease.

a. Gangwar, S.P., Green, M.N., Michard, E., Simon, A.A., Feijó, J.A., & Sobolevsky, A.I. (2020). Structure of the Arabidopsis Glutamate Receptor-like Channel GLR3.2 Ligand-Binding Domain. *Structure*.doi:10.1016/j.str.2020.09.006
b. Green, M. N., Gangwar, S. P., Michard, E., Simon, A. A., Portes, M. T., Barbosa-Caro, J., Wudick, M.M., Lizzo, M.A., Klykov, O., Yelshanskaya, M.V., Feijó, J.A.,& Sobolevsky, A. I. (2021). Structure of the Arabidopsis thaliana glutamate receptor-like channel GLR3. 4. *Molecular Cell*.

D. Additional Information: Research Support and/or Scholastic Performance

The grading scale for all the undergraduate institutions: 4.0 GPA scale (meaning A+ is equal points as an A).

YEAR	COURSE TITLE	GRADE
	PALOMAR COLLEGE	
2008	Hip Hop I (DANC 155)	A
2008	Hawaiian And Tahitian Dance I (DANC 158)	A
2008	Fundamentals of Ballet	A
2008	Power Reading (READ 110)	A
2008	Introduction to Sociology (SOC 100)	A
2010	Elementary German I (GERM 101)	A
2010	College Algebra (MATH 110)	B
2010	Elementary German II (GERM 102)	A
2010	Oral Communication (SPCH 100)	A
2010	MGMT of Speech Activities (SPCH 145)	A
2010	Practical Public Speaking (SPCH 160)	A
2011	General Biology (BIOL 100)	A
2011	Principles of Economics (Micro) (ECON 102)	A
2011	German III (GERM 201)	A
	MIRACOSTA COLLEGE	
2011	Computer Applications (CSIT 110)	A
2011	Sensory Analysis of Wines (HORT 145)	A
2011	Trigonometry (MATH 130)	P
2011	Pre-Calculus Math (MATH 135)	P
2012	Composition and Reading (ENGL 100)	A
2012	Preparatory Chemistry (CHEM108)	B
2012	Western Civ Since 1648 (Hon) (HIST 104H)	A
2012	Calculus & Analytical Geometry I (MATH 150)	B
2012	General Chemistry (CHEM 110)	A
2012	Fundamentals of Microbiology (BIO 230)	B
2013	Critical thinking, Composition & Literature (ENGL 201)	A
2013	General Chemistry (CHEM 111)	A
2013	Calculus & Analytical Geometry II (MATH 155)	A
2013	Statistics for Behavioral Sci (SOC 104)	A
2013	Renaissance/Modern Art (ART 259)	A
2013	Critical Thinking, Comp and Lit (ENGL 201)	A
2013	Evolution, Biodiversity, Organismal (BIO 202)	A
2013	Organic Chemistry I (CHEM 210)	A
2013	Intro Physics I (PHYS 111)	A
2014	Biochemistry, Cell, Gene, Molecular (BIO 204)	A

YEAR	COURSE TITLE	GRADE
2014	Organic Chemistry II (CHEM 211)	A
2014	Principles of Physics II (PHYS 152)	A
2014	General Psychology (PHYC 101)	P
	UC SAN DIEGO	
2014	Genetics (BICD 100)	B
2014	Biochemical Structure & Function (CHEM 114A)	A-
2014	Intermediate German I (LTGM 2A)	A
2014	Recombinant DNA Techniques (BIMM101)	W
2015	Biochem Energetics & Metabolism (CHEM 114B)	A
2015	Intermediate German II (LTGM 2B)	A
2015	Calculus & Analyt Geom/Sci&Engnr (MATH 20C)	A+
2015	Biosynthesis of Macromolecules (CHEM 114C)	C
2015	Intermediate German III (LTGM 2C)	A
2015	Intro/Differential Equations (MATH 20D)	A-
2015	Protein Biochemistry Lab (CHEM 108)	B+
2015	Inorganic Chemistry I (CHEM 120A)	B
2015	Physics-Flu, Wav, Thrmdyn, Optics (PHYS 2C)	B-
2016	Recombinant DNA Laboratory (CHEM 109)	A-
2016	Physical Chem: Quantum Mech (CHEM 126)	B
2016	Drug Synthesis and Design (CHEM 168)	B+
2016	Analytical Chemistry Lab (CHEM 100A)	B
2016	Physical Chem: Thermodynamics (CHEM 127)	A-
2016	Chemistry Internship (CHEM 197)	P
2016	Language, Culture & Education (SOCI 117)	P
2016	Philosophy and Race (PHIL 170)	P
2016	E. Asia Thought/Comp Perspectv (POLI 113A)	P
2017	Physical Chemistry Laboratory (CHEM 105A)	C
2017	Pharmacology and Toxicology (CHEM 118)	C+
2017	Physics-Mechanics (PHYS 2A)	C
	COLUMBIA UNIVERSITY	
2017	Biochem/Molecular/Cell Bi (BCHM 6300 G)	B
2017	Doctoral Research in Nutrition (NUTR 9011 G)	P
2017	Doctoral Seminar in Nutrition (NUTR 9205 G)	P
2017	Mechanisms in Hum Disease (PATH 6003 G)	A-
2018	Resp Cond of Res/Rel Plcy (CMBS 4010 G)	P
2018	Biochem, Cell/Molecular Bi (CMBS 6301 G)	B+
2018	Molecular/Cell Bio of Nutr (NUTR 4020 G)	A-
2018	Doctoral Research in Nutr (NUTR 9011 G)	P
2018	Doctoral Seminar in Nutri (NUTR 9205 G)	P
2018	Mechanisms in Hum Disease (Path 6004 G)	A-
2018	Intro-Biostatistical Meth (BIST 6104 P)	B
2018	Doctoral Seminar in Nutri (NUTR 9205 G)	P
2018	Reviews in Nutrition (NUTR 9300 G)	P
2019	Doctoral Seminar in Nutri (NUTR 9205 G)	P
2019	Reviews in Nutrition (NUTR 9205 G)	P
2020	Doctoral Seminar in Nutri (NUTR 9205 G)	P
2020	Reviews in Nutrition (NUTR 9300 G)	P
2020	Grants & Contracts (MEDI 9780 M)	P
2020	Cryo-Electron Microscopy (BCHM 6400 GR)	A

BIOGRAPHICAL SKETCH

NAME: Gangwar, Shanti Pal

eRA COMMONS USERNAME (credential, e.g., agency login): SPGANGWA

POSITION TITLE: Post-Doctoral Research Scientist

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Start Date MM/YYYY	Completion Date MM/YYYY	FIELD OF STUDY
Kumaun University, India	MSc	July 2005	June 2007	Biotechnology
Jawaharlal Nehru University, India	PhD	July 2007	Feb 2014	Biophysics/Structural Biology
University of Texas Medical Branch (UTMB), USA	Postdoc	Sep 2014	Jan 2019	Biophysics/Structural Biology/Neuroscience
Columbia University, USA	Postdoc	March 2019	Current	Biophysics/Structural Biology/Neuroscience

A. Personal Statement

My long-term goal is to understand the structural and functional perspective of ion channels involved in neuronal communication and several neurological disorders including Alzheimer's disease, amyotrophic lateral sclerosis, epilepsy, and ischemia. My academic and research training has provided me skills in biophysics, X-ray crystallography, molecular biology, biochemistry, and Cryo-EM. As a graduate student, I learned X-ray crystallography studying transcript factors from human and *Mycobacterium tuberculosis*. As a postdoc under the guidance of **Dr. Gabrielle Rudenko at UTMB,** I was introduced to the scope and importance of neurodevelopmental/neuropsychiatric disorders and I started studying on synaptic proteins. Subsequently, my research findings on the structural and mechanistic details of synapse adhesion molecules (Neuroligin-MDGA1) have suggested strategies to design structure-guided peptides/small molecules modulating protein-protein interactions as therapeutics for neurodevelopmental disorders. Now I am advancing my research further by studying the neuronal ion channels under the supervision of **Dr. Alexander Sobolevsky at Columbia University**. The proposed research on the structural studies of the ionotropic glutamate receptors and its outcomes would expand our understanding of the gating mechanism as well as modulation by small molecules/inhibitor to develop therapeutics. The proposed research outlines a set of career developmental activities such as grant writing, public speaking, management in the lab, mentoring students, and altogether enhancing my abilities in becoming a successful independent investigator. Given the competitive nature of the proposed research area, it will be an excellent opportunity for me to work on the structure-function relationship of ion channels.

- **Gangwar, SP*.,** Green, M.N*., Michard, E*., Simon, A.A., Feijo, J.A., and Sobolevsky, A.I. (2019) Structure of the *Arabidopsis* Glutamate Receptor-like Channel GLR3.2 Ligand-Binding Domain. Structure. https://doi.org/10.1016/j.str.2020.09.006 **(Cover - page)**

B. Honors and Awards

- Awarded **Best Poster** Prize in a poster presentation at *23rd Annual Sealy Center for Structural Biology Symposium*, 28th April 2018, University of Texas Medical Branch, Texas USA.

- Awarded **Best Poster** Prize in a poster presentation at a 4th International Symposium on "*Recent trends in Macromolecular Structure and Function"*, Jan 21-23, 2010, University of Madras, Chennai, India.

- Awarded **Junior Research Fellowship** and **Senior Research Fellowship** from Council of Scientific and Industrial Research (CSIR) and University Grants Commission (UGC) India and Qualified Graduate Aptitude Test in Engineering **(GATE)** 2006 by Department of Higher Education, MHRD, Government of India.

C. Positions and Employment

2014 - 2019 Postdoctoral Researcher, University of Texas Medical Branch, Galveston, TX
2019 - current Postdoctoral Researcher, Columbia University, New York, NY

D. Contributions to Science

Graduate Career

My graduate research contributions focused on the transcription factors from human and *Mycobacterium tuberculosis*. The outcomes of the research were of significate importance that provided insights that full-length Erg is a highly nonglobular protein, which is subjected to DNA binding autoinhibition mechanism. The DNA binding domain (ETS domain) of human Erg is a winged helix-turn-helix and binds to DNA using its particular helix. Modulation of this DNA-Protein interaction by small molecules/peptides may open up new therapeutic avenues in the field of prostate cancer.

The *Mycobacterium tuberculosis* transcriptional regulator EspR contains an N-terminal helix–turn–helix DNA binding domain and a C-terminal dimerization domain. Structural study and comparison of EspR in different crystal forms indicated that the N-terminal helix–turn–helix domain of EspR acquires a rigid structure in different crystal forms. However, significant structural differences were observed in the C-terminal domain of EspR. The interaction, stabilization energy and buried surface area analysis of EspR in the different crystal forms have provided information about the physiological dimer interface of EspR.

Sharma, R., **Gangwar, SP.,** Saxena, A.K. (2018) Comparative structure analysis of the ETSi domain of ERG3 and its complex with the E74 promoter DNA sequence. *Acta Crystallogr. Section F Biol. Crystallogr*.F74. 656-663.

Gangwar, S. P., Meena, S. R. and Saxena, A. K. (2014). Comparison of four different crystal forms of Mycobacterium tuberculosis ESX-1 secreted protein regulator, EspR. *Acta Crystallogr. Section F Biol. Crystallogr*.F70.

Gangwar, S. P., Dey, S., and Saxena, A. K. (2012). Structural modeling and DNA binding auto-inhibition analysis of Ergp55, a critical transcription factor in prostate cancer. *PLoS ONE* 7(6), e39850.

Postdoctoral Career

During my postdoc at UTMB, Texas, I have studied the structural perspective of the synapse-related, organizers/adhesion, proteins critical in brain development using X-ray crystallography, and other biophysical methods. The outcomes of this research focus on how a synapse organizer, MDGA1, interacts with Neuroligin and regulates the interaction with Neurexin and Neuroligin. On the basis of this structural information, we designed small peptides modulating the Neuroligin and MDGA1 interaction and tested their efficacy by related biophysical methods. The next goal of this project is to increase the binding affinity of these peptides by optimizing the peptide sequence making them protease-resistant and then test *in vivo* / in animal models to explore the therapeutic potential to recalibrate excitation-inhibition imbalances at the synapse.

- Fan, S**., Gangwar, SP.,** Machius, M., and Rudenko, G. (2020) Interplay between hevin, SPARC, and MDGAs: modulators of neurexin-neuroligin trans-synaptic bridges. **Structure,** https://doi.org/10.1016/j.str.2021.01.003

- **Gangwar, SP.,** Zhong, X., Seshadrinathan, S., Chen, H., Machius, M., and Rudenko, G. (2017). Molecular Mechanism of MDGA1: Regulation of Neuroligin 2: Neurexin Trans-synaptic Bridges**.** ***Neuron.*** 2017 Jun 21;94(6):1132-1141.e4.

- Kim, M.J., Biag, J., Fass, D.M., Lewis, M.C., Zhang, Q., Fleishman, M., **Gangwar, S.P.**, Machius, M., Fromer, M., Purcell, S.M., Premont, R.T., McCarroll, S.A., Rudenko, G., Scolnick, E.M., Haggarty, S.J. Functional analysis of rare variants found in schizophrenia implicates a critical role for GIT1-PAK3 signaling in neuroplasticity. ***Molecular Psychiatry***.2017, 22(3):417-429.

D. Additional Information: Research Support and/or Scholastic Performance

Publications

1. Green, MN*., **Gangwar, SP*.,** Michard, E., Simon, AA., Portes, MT., Barbosa-Caro, J., Wudick, MM., Lizzio, MA., Klykov, O., Yelshanskaya, MV., Feijo, JA., and Sobolevsky, AI. (2021) Structure of the Arabidopsis thaliana Glutamate Receptor-Like Channel GLR3.4. **Mol Cell** https://doi.org/10.1016/j.molcel.2021.05.025 (equal contribution)

2. Fan, S., **Gangwar, SP.,** Machius, M., and Rudenko, G. (2020) Interplay between hevin, SPARC, and MDGAs: modulators of neurexin-neuroligin trans-synaptic bridges. **Structure,** https://doi.org/10.1016/j.str.2021.01.003

3. **Gangwar, SP.,** Bandyopadhyay, A., and Saxena, AK. (2020) Structural studies on *M. tuberculosis* decaprenyl phosphoryl-β-D-ribose epimerase-2 enzyme involved in cell wall biogenesis. **BioRxiv.** doi**:** https://doi.org/10.1101/2020.10.15.341941

4. **Gangwar, SP.,** Green, M.N., Michard, E., Simon, A.A., Feijo, J.A., and Sobolevsky, A.I. (2019) Structure of the *Arabidopsis* Glutamate Receptor-like Channel GLR3.2 Ligand-Binding Domain. **Structure***.* https://doi.org/10.1016/j.str.2020.09.006 **(Cover - page)**

5. Sharma, R., **Gangwar, SP.,** Saxena, A.K. (2018) Comparative structure analysis of the ETSi domain of ERG3 and its complex with the E74 promoter DNA sequence. ***Acta Crystallogr. Section F Biol. Crystallogr.***F74. 656-663.

6. **Gangwar, SP.,** Zhong, X., Seshadrinathan, S., Chen, H., Machius, M., and Rudenko, G. (2017). Molecular Mechanism of MDGA1: Regulation of Neuroligin 2: Neurexin Trans-synaptic Bridges**.** ***Neuron****.* 2017 Jun 21;94(6):1132-1141.e4.

7. Kim, M.J., Biag, J., Fass, D.M., Lewis, M.C., Zhang, Q., Fleishman, M., **Gangwar, S.P.**, Machius, M., Fromer, M., Purcell, S.M., Premont, R.T., McCarroll, S.A., Rudenko, G., Scolnick, E.M., Haggarty, S.J. Functional analysis of rare variants found in schizophrenia implicates a critical role for GIT1-PAK3 signaling in neuroplasticity. ***Molecular Psychiatry*****.** 2017, 22(3):417-429.

8. **Gangwar, S. P.,** Meena, S. R. and Saxena, A. K. (2014). Comparison of four different crystal forms of Mycobacterium tuberculosis ESX-1 secreted protein regulator, EspR. ***Acta Crystallogr. Section F Biol. Crystallogr***.F70.

9. **Gangwar, S. P.,** Meena, S. R. and Saxena, A. K. (2014). Structure of the carboxy-terminal domain of *M. tuberculosis* CarD protein: an essential rRNA transcriptional regulator. ***Acta Crystallogr. Section F Biol. Crystallogr****. F70.*

10. **Gangwar, S. P.,** Dey, S., and Saxena, A. K. (2012). Structural modeling and DNA binding auto-inhibition analysis of Ergp55, a critical transcription factor in prostate cancer. ***PLoS ONE*** 7(6), e39850.

11. **Gangwar, S. P.,** Meena, S. R., and Saxena, A. K. (2012). Purification, crystallization and preliminary X-ray crystallographic analysis ETS domain of Ergp55 in complex with *c-fos* promoter DNA sequence. ***Acta Crystallogr. Section F Biol. Crystallogr****. F68,* 1333- 1336.

12. Meena, S. R., **Gangwar, S. P.** and Saxena A. K. (2012). Purification, crystallization and preliminary X-ray crystallographic analysis of ATPase domain of TAP in nucleotide-free, ADP, vanadate and azide inhibited form. ***Acta Crystallogr. Section F Biol. Crystallogr. F****68,* 655-658

13. **Gangwar, S. P.,** Meena, S. R. and Saxena, A. K. (2011). Cloning, purification, crystallization and preliminary X-ray analysis of EspR: a secreted transcription factor from *M. tuberculosis*. ***Acta Crystallogr. Section F Biol. Crystallogr****. F67*, 83-86.

Symposia/Conference paper presentations

- **Gangwar SP,** Zhong X, Seshadrinathan S, Chen H, Machius M and Rudenko G. (2017) Structural Insights Into The Regulation Of Neuroligin2: Neurexin Trans-Synaptic Bridge By MDGA1. 27th Annual Keck Center Research Conference: *Innovations in Interdisciplinary Neuroscience,* October 27, 2017, Houston, Texas.

- **Gangwar, S. P. (**2013) Structural and functional dissection of the human Ergp55 oncoprotein.*42nd National seminar on crystallography and International workshop on the application of X-ray diffraction for drug discovery*, November 21-23, New Delhi- India.
- **Gangwar, S. P.,** Meena, S. R. and Saxena, A. K. (2012). Structure and functional analysis of key proteins involved in Mtb ESX-1 protein export pathway: potential drug targets. *National symposium of microbes in Health and Agriculture,* March 12-13, JNU, New Delhi, India. **(Best poster presentation award).**

- Meena, S. R., **Gangwa**r, **S. P.,** and Saxena, A. K. (2010) Elucidation of the mechanism of ATP hydrolysis cycle of the Transporter Associated with Antigen Processing (TAP). *4th International Symposium on Recent Trends in Macromolecular Structure and Function,* January21-23, Chennai, India. **(Best poster presentation award)**.

- Meena, S. R., **Gangwar, S. P.** and Saxena, A. K. (2008) Structure analysis of Transporter Associated with Antigen Processing (TAP). *International Symposium on Novel Strategies for Targeted Prevention and Treatment of Cancer*, JNU, India.

- **Gangwar, S. P.,** Meena, S. R., and Saxena, A. K. (2008) Structure analysis of ERG oncoprotein: A potential target to develop a prostate cancer drug. *International Symposium on Novel Strategies for Targeted Prevention and Treatment of Cancer,* JNU, India.

Skills and Techniques

- **Protein expression and Purification:** Membrane protein as well as soluble protein from *E. coli*, Insect cell line, and Mammalian cells.

- **Macromolecular Crystallography:** Crystallization of proteins and protein-DNA complexes. Crystal handling and mounting, Crystal soaking, Data collection at home source and synchrotron (BM14 beamline at ESRF, LS-CAT, 19ID SBC-CAT at APS). Data processing using iMOSFLM and HKL2000. Structure determination by experimental Phasing (using Selenomethionine and Iodide, SAD) and Molecular replacement using CCP4 suite and Phenix suite, structure visualization, and refinement using coot.

- **Cryo-EM:** Sample preparation, Vitrobot handling, data processing using Relion and CryoSparc

- **Techniques:** Genomic and plasmid DNA isolation, PCR, Gene cloning. Affinity, size exclusion, ion exchange Chromatography, Western blotting, Dot-blot, CD spectroscopy, Fluorescence polarization assay, ITC, Protein thermal shift assay, SPR, BLAST, ClustalW, Modweb and Swiss-Model, Molecular dynamics simulation using Gromacs and structure visualization using Pymol.

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
Follow this format for each person. DO NOT EXCEED FIVE PAGES.

NAME: Sobolevsky, Alexander

eRA COMMONS USER NAME (credential, e.g., agency login): SOBOLEVS

POSITION TITLE: Associate Professor

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE (if applicable)	END DATE MM/YYYY	FIELD OF STUDY
Moscow Inst. of Physics and Technology, Moscow	MS	06/1996	Bioengineering
Moscow Inst. of Physics and Technology, Moscow	PHD	11/1999	Biophysics
Stony Brook University, NY	Post-Doc	08/2004	Neurobiology
Columbia University, NY/Vollum Institute, OHSU, OR	Post-Doc	08/2010	Structural Biology

A. Personal Statement

My lab studies structure and function of ion channels, including ionotropic glutamate receptors (iGluRs) and transient receptor potential (TRP) channels, using a combination of biochemical and biophysical methods and cryo-electron microscopy (cryo-EM) in particular. I have an expertise in solving structures of integral membrane proteins by both X-ray crystallography and cryo-EM and an extensive experience in using methods of characterizing ion channels function, including patch-clamp, double-electrode voltage-clamp recordings and Fura-2-based ratiometric fluorescent measurements of intracellular calcium. I also have an expertise in analyzing different types of ion channel inhibition using a combination of electrophysiology, protein engineering and kinetic modeling. With such expertise and experiences, I studied the mechanisms of ionotropic glutamate receptor (iGluR) inhibition by ion channel blockers, including the only FDA-approved NMDA receptor channel blocker Memantine, currently used for treatment of Alzheimer's disease. I solved the first full length crystal structure of ionotropic glutamate receptor. My lab solved numerous structures of full-length iGluRs, including the first agonist-bound, open and desensitized state structures and proposed the first complete structural model of iGluR gating. Using X-ray crystallography, my lab determined the structural mechanism of iGluR inhibition by noncompetitive inhibitors, including Perampanel that is currently used for treatment of epilepsy. My lab also solved the first TRP channel crystal structure (the structure of TRPV6). Using cryo-EM, my lab determined structures of human TRPV6 in different conformations and proposed the mechanism of TRPV6 activation. Similarly, my lab solved the first TRPV3 structure and structures of TRPV3 in different conformations and proposed the mechanism of ligand-induced TRPV3 activation. Recently, we solved structures of TRPV3 in temperature-dependent closed, intermediate and open states, which for the first time uncovered the structural bases of TRP channel activation by temperature. As a result of my previous experiences, I am aware of the importance of frequent communication among project members and of constructing a realistic research plan, timeline, and budget. My current research plans build logically on my prior work.

Ongoing and recently completed projects that I would like to highlight include:

R01 CA206573
Sobolevsky (PI)
01/12/2017-12/31/2021
Structure and Function of Transient Receptor Potential channels

R01 NS083660
Sobolevsky/Kurnikova (MPI)
09/30/2013-06/30/2023
Structure and Function of AMPA subtype ionotropic glutamate receptors

R01 NS107253
Sobolevsky (PI)
08/01/2018-05/31/2023
Single-particle cryo-EM characterization of AMPA receptor functional states

NSF 1818213
Sobolevsky/Kurnikova/Stern-Bach (MPI)
08/01/2018-07/31/2022
Collaborative Research: Towards development of the structural determinants of the Glutamate receptor gating regulation by auxiliary membrane anchored proteins

Pew Scholar Award
Sobolevsky (PI)
07/01/2013-07/31/2018
Structure and Function of TRP Channels

Irma Hirschl Career Scientist Award
Sobolevsky (PI)
01/01/2015-12/31/2019
Molecular mechanisms of ionotropic glutamate receptor gating, assembly and regulation

Citations:

1. McGoldrick LL, Singh AK, Saotome K, Yelshanskaya MV, Twomey EC, Grassucci RA, Sobolevsky AI. Opening of the human epithelial calcium channel TRPV6. **Nature**. 2018 Jan 11;553(7687):233-237. PubMed Central PMCID: PMC5854407.
2. Twomey EC, Yelshanskaya MV, Grassucci RA, Frank J, Sobolevsky AI. Channel opening and gating mechanism in AMPA-subtype glutamate receptors. **Nature**. 2017 Sep 7;549(7670):60-65. PubMed Central PMCID: PMC5743206.
3. Saotome K, Singh AK, Yelshanskaya MV, Sobolevsky AI. Crystal structure of the epithelial calcium channel TRPV6. **Nature**. 2016 Jun 23;534(7608):506-11. PubMed Central PMCID: PMC4919205.
4. Yelshanskaya MV, Li M, Sobolevsky AI. Structure of an agonist-bound ionotropic glutamate receptor. **Science**. 2014 Aug 29;345(6200):1070-4. PubMed Central PMCID: PMC4383034.

B. Positions, Scientific Appointments and Honors

Positions and Scientific Appointments

2017 -	Associate Professor, Columbia University, New York, NY
2010 - 2017	Assistant Professor, Columbia University, New York, NY
2005 - 2010	Post-doctoral Research Fellow, Vollum Institute, Oregon Health and Science University, Portland, OR
2004 - 2005	Post-doctoral Research Fellow, Columbia University, New York, NY
2000 - 2004	Post-doctoral Research Fellow, Stony Brook University, Stony Brook, NY
1996 - 1999	Pre-doctoral Research Fellow, Moscow Institute of Physics and Technology, Moscow
1993 - 1996	Pre-diploma Research Fellow, Moscow Institute of Physics and Technology, Moscow

Honors

2017	Amgen Young Investigator Award, Amgen
2015	Irma T. Hirschl Career Scientist Award, Irma T. Hirschl Trust
2013	Pew Scholar Award, Pew Charitable Trusts
2012	Schaefer Research Scholar Award, Dr. Ludwig Schaefer Fund
2011	Klingenstein Fellowship Award in the Neurosciences, Esther A. & Joseph Klingenstein Fund
2002	Postdoctoral Travel Award for participation in the 32nd Annual Meeting of the Society for Neuroscience, Burroughs Wellcome Fund

2000	Travel Grant for participation in the 31st Annual Meeting of the Society for Neuroscience, International Brain Research Organization
1999	International Soros Science Education Program Grant, Soros Foundation
1998	International Soros Science Education Program Grant, Soros Foundation
1998	Travel Grant for participation in the 29th Annual Meeting of the Society for Neuroscience, International Brain Research Organization

C. Contribution to Science

1. N-methyl-D-aspartate (NMDA) receptors are a subtype of ionotropic glutamate receptors that is critical to neuronal development and synaptic plasticity, associated with memory formation and learning and implicated in acute and chronic neuronal death, associated with brain trauma and neurological disorders. Ion channel blockers of NMDA receptors therefore have an enormous drug potential. We have been among the first research groups to study the mechanism of ion channel block of NMDA receptors by various derivatives of aminoadamantane, one of which, Memantine (NAMENDA), have become the first and so far the only drug acting at NMDA receptors that has been approved by FDA for treatment of moderate to severe Alzheimer's disease. We developed a set of new kinetic criteria to analyze the mechanism of blocker interaction with ion channel gating machinery. Using this set, we were the first to discover that Mg2+ interacts with NMDA receptors via the trapping block mechanism. The discovery of the trapping block of NMDA receptor channels by Mg2+ led to reevaluation of the role of Mg2+ and NMDA receptors in neurotransmission across excitatory synapses in the brain.

    a. Sobolevsky AI, Yelshansky MV. The trapping block of NMDA receptor channels in acutely isolated rat hippocampal neurones. **J Physiol**. 2000 Aug 1;526 Pt 3:493-506. PubMed Central PMCID: PMC2270033.
    b. Sobolevsky AI, Koshelev SG, Khodorov BI. Probing of NMDA channels with fast blockers. **J Neurosci**. 1999 Dec 15;19(24):10611-26. PubMed Central PMCID: PMC6784965.
    c. Sobolevsky AI, Koshelev SG, Khodorov BI. Interaction of memantine and amantadine with agonist-unbound NMDA-receptor channels in acutely isolated rat hippocampal neurons. **J Physiol**. 1998 Oct 1;512 (Pt 1):47-60. PubMed Central PMCID: PMC2231181.
    d. Sobolevsky A, Koshelev S. Two blocking sites of amino-adamantane derivatives in open N-methyl-D-aspartate channels. **Biophys J**. 1998 Mar;74(3):1305-19. PubMed Central PMCID: PMC1299478.

2. Before the structures of the full length iGluR become available, one could only guess what are the structural organization of the iGluR channel and the mechanisms of pore opening and closure. To gain insights into the structure of the NMDA receptor ion channel pore and the structural rearrangements during gating, we used the substituted cysteine accessibility method (SCAM). The NMDA receptor is an obligate heterotetramer composed of two or more different subunits. We individually mutated residues in the transmembrane portion of the two major subtypes of NMDA receptor subunits, NR1 and NR2. We identified the boundaries and the pore-facing surfaces of the transmembrane domains, their relative contribution to the ion channel pore and gating and the amino acid residues in the pore involved into receptor activation and desensitization as well as binding of the channel blockers. We were among the first to discover the asymmetrical contribution of the NR1 and NR2 subunits to channel pore structure and gating and the central role of the M3 segment in NMDA receptor gating.

    a. Sobolevsky AI, Prodromou ML, Yelshansky MV, Wollmuth LP. Subunit-specific contribution of pore-forming domains to NMDA receptor channel structure and gating. **J Gen Physiol**. 2007 Jun;129(6):509-25. PubMed Central PMCID: PMC2151626.
    b. Wollmuth LP, Sobolevsky AI. Structure and gating of the glutamate receptor ion channel. **Trends Neurosci**. 2004 Jun;27(6):321-8. PubMed PMID: 15165736.
    c. Sobolevsky AI, Rooney L, Wollmuth LP. Staggering of subunits in NMDAR channels. **Biophys J**. 2002 Dec;83(6):3304-14. PubMed Central PMCID: PMC1302406.
    d. Sobolevsky AI, Beck C, Wollmuth LP. Molecular rearrangements of the extracellular vestibule in NMDAR channels during gating. **Neuron**. 2002 Jan 3;33(1):75-85. PubMed PMID: 11779481.

3. We used SCAM and patch-clamp recordings to study structure and function of homotetrameric AMPA subtype iGluRs. We identified pore-forming elements and residues involved in AMPA receptor gating. We discovered mutations outside the ligand binding domain (LBD) – in the linkers connecting the LBD to the ion channel – that resulted in either enhancement or nearly complete oblation of AMPA receptor desensitization. We found that AMPA receptors are unique compared to other tetrameric ion channels and that despite the subunit assembly is homomeric, contribution of individual subunits to the ion channels pore is different leading to the overall two- rather than four-fold rotation symmetry of the ion channel in the active state.

    a. Sobolevsky AI, Yelshansky MV, Wollmuth LP. State-dependent changes in the electrostatic potential in the pore of a GluR channel. **Biophys J**. 2005 Jan;88(1):235-42. PubMed Central PMCID: PMC1305001.
    b. Yelshansky MV, Sobolevsky AI, Jatzke C, Wollmuth LP. Block of AMPA receptor desensitization by a point mutation outside the ligand-binding domain. **J Neurosci**. 2004 May 19;24(20):4728-36. PubMed Central PMCID: PMC6729461.
    c. Sobolevsky AI, Yelshansky MV, Wollmuth LP. The outer pore of the glutamate receptor channel has 2-fold rotational symmetry. **Neuron**. 2004 Feb 5;41(3):367-78. PubMed PMID: 14766176.
    d. Sobolevsky AI, Yelshansky MV, Wollmuth LP. Different gating mechanisms in glutamate receptor and K+ channels. **J Neurosci**. 2003 Aug 20;23(20):7559-68. PubMed Central PMCID: PMC6740752.

4. The transient receptor potential (TRP) channels are a superfamily of cation permeable ion channels that are widely known for their role as transducers of sensory modalities, including temperature, taste, olfaction, vision, hearing and touch. TRP channels are also crucial for a diverse range of physiological processes, such as neurite outgrowth, hormone secretion and control of vascular tone. Accordingly, mutations or malfunction of TRP channels are associated with numerous human diseases, including cardiovascular, renal, nociceptive and metabolic disorders. We solved the first crystal structure of TRP channel, Ca2+-selective channel TRPV6 that plays vital roles in calcium homeostasis as a Ca2+ uptake channel in epithelial tissues and is implicated in development and progression of numerous forms of cancer. We also determined the structural bases of TRPV6 allosteric regulation and calcium-induced calmodulin-mediated inactivation. We also solved the first structure of TRPV3 and determined structural beses of TRPV3 activation. Our results provide a structural foundation to understand the regulation of epithelial Ca2+ uptake and its role in pathophysiology and provide information necessary for drug design.

    a. Singh AK, McGoldrick LL, Sobolevsky AI. Structure and gating mechanism of the transient receptor potential channel TRPV3. **Nat Struct Mol Biol**. 2018 Sep;25(9):805-813. PubMed Central PMCID: PMC6128766.
    b. Singh AK, Saotome K, McGoldrick LL, Sobolevsky AI. Structural bases of TRP channel TRPV6 allosteric modulation by 2-APB. **Nat Commun**. 2018 Jun 25;9(1):2465. PubMed Central PMCID: PMC6018633.
    c. McGoldrick LL, Singh AK, Saotome K, Yelshanskaya MV, Twomey EC, Grassucci RA, Sobolevsky AI. Opening of the human epithelial calcium channel TRPV6. **Nature**. 2018 Jan 11;553(7687):233-237. PubMed Central PMCID: PMC5854407.
    d. Saotome K, Singh AK, Yelshanskaya MV, Sobolevsky AI. Crystal structure of the epithelial calcium channel TRPV6. **Nature**. 2016 Jun 23;534(7608):506-11. PubMed Central PMCID: PMC4919205.

5. High resolution structural information about ionotropic glutamate receptors opens new horizons to understanding their gating mechanism and regulation at the molecular level as well as makes iGluRs a novel pharmacological platform for characterizing new compounds with diverse activities for use as therapies in neurological diseases. My lab has solved the first crystal structure of the full length AMPA receptor in complex with agonist, crystallographically discovered novel binding sites of antiepileptic drugs, obtained the first cryo-EM structures of AMPA receptor complexes with the auxiliary subunits stargazin and GSG1L, and solved the first structures of AMPA receptor in the open and desensitized states.

a. Twomey EC, Yelshanskaya MV, Grassucci RA, Frank J, Sobolevsky AI. Channel opening and gating mechanism in AMPA-subtype glutamate receptors. **Nature**. 2017 Sep 7;549(7670):60-65. PubMed Central PMCID: PMC5743206.
b. Yelshanskaya MV, Singh AK, Sampson JM, Narangoda C, Kurnikova M, Sobolevsky AI. Structural Bases of Noncompetitive Inhibition of AMPA-Subtype Ionotropic Glutamate Receptors by Antiepileptic Drugs. **Neuron**. 2016 Sep 21;91(6):1305-1315. PubMed Central PMCID: PMC5033713.
c. Twomey EC, Yelshanskaya MV, Grassucci RA, Frank J, Sobolevsky AI. Elucidation of AMPA receptor-stargazin complexes by cryo-electron microscopy. **Science**. 2016 Jul 1;353(6294):83-6. PubMed Central PMCID: PMC5125255.
d. Yelshanskaya MV, Li M, Sobolevsky AI. Structure of an agonist-bound ionotropic glutamate receptor. **Science**. 2014 Aug 29;345(6200):1070-4. PubMed Central PMCID: PMC4383034.

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