BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
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NAME: Ortlund, Eric A.

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

POSITION TITLE: Associate Professor, Department of Biochemistry

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)*	Completion Date MM/YYYY	FIELD OF STUDY
University of South Carolina	B.S.	05/1997	Chemistry
University of South Carolina	Ph.D.	05/2002	Chemistry, Biochemistry
University of North Carolina – Chapel Hill	Post-Doc	06/2007	Biochemistry Structural Biology

A. Personal Statement

Our lab is interested in the structural and biochemical mechanism by which human nuclear receptors and lipid binding proteins influence in stress, development, and lipid metabolism. I have a strong track record of collaboration, which has led to numerous contributions to both the nuclear receptor field and the lipid signaling community. Over the past 10 years, we have become interested not only in how lipids directly activate transcription factors but how they are trafficked to the nucleus by soluble lipid shuttling proteins. We have focused on the liver receptor homologue 1, LRH-1, which is a phospholipid-regulated nuclear receptor that controls bile acid, cholesterol, and lipid homeostasis. In collaboration with Dr. David Moore (Baylor, TX), we have identified *bona fide* phospholipid agonists for this receptor and have shown that these novel ligands possess powerful antidiabetic properties stemming in part from their ability to lower hepatic triglyceride levels. We are now focused on imaging LRH-1-mediated transcriptional complexes using cryoEM. Nuclear receptors have no intrinsic enzymatic activity and function by recruiting transcriptional coregulator to chromatin. Therfore, to understand how ligands are driving NR-mediated gene expression we must image these complexes. cryoEM is the only available tool for this effort and we have collected exciting preliminary data on a stable and monodisperse NR-coregulator complexes on DNA. This exciting preliminary work enables the proposed cryoEM studies and future mechanistic studies centered on the ligand-regulated LRH-1 transcriptome. Emory has made large investments to build infrastructure and expertise in cryoEM. We are leveraging these resources by taking advantage one-on-one training and cryoEm workshops available through our core (and nationally). We hold weekly meetings with the cryoEM core and other four labs on campus using these techniques and host bi-weekly presentations where students and trainees present their current research centered on single particle cryoEM or tomographic techniques. To achieve high-resolution, we desperately need access to the resources and expertise offered by the NCCAT!

B. Positions and Honors

Positions and Employment

2002-02 Postdoctoral Fellow, Laboratory of Lukasz Lebioda; Dept of Chemistry, University of South Carolina, Columbia, SC
2003-07 Postdoctoral Fellow, Laboratory of Matthew Redinbo; Department of Chemistry, University of North Carolina, Chapel Hill; Lineberger Comprehensive Cancer Center, UNC
2007-present Associate Professor; Department of Biochemistry, Emory University, Atlanta, GA
2009-present Member, Executive Committee, Molecular Systems Pharmacology Graduate Program

2016-2018 Founder and Director of Emory Integrated Lipidomics Core

2019-Present Founder and Director of Emory Integrated Metabolomics and Lipidomics Core

Other Experience and Professional Memberships

2010-14 PC4 and PC2 study section, American Heart Association
2012 RFA study section NIH Blood and Vascular Branch, National Heart, Lung, and Blood Institute, NIH
2012-13 French National Research Agency Grant Reviewer
2012-13 Foundation for Polish Science Grant Reviewer
2014 NIH Molecular and Cellular Endocrinology study section, NIH, *Ad hoc*
2012- Editorial Board, ASPET, Molecular Pharmacology
2012- Reviewer Board, Journal of Pediatric Biochemistry
2013- Editorial Board, Nuclear Receptor Research
2013- Editorial Board, Hepatology
2015 NIH LCMI Special Emphasis Panel/Scientific Review Group
2015 NIH Special Emphasis Panel ZRG1 EMNR-R(56), Cellular Mechanisms of Metabolism and Obesity
2016 Austrian Science Fund, study section
2016- NIH Molecular and Cellular Endocrinology study section, Chair
2017 NIH ZRG1 EMNR A 07, Molecular and Cellular Endocrinology, Chair

Honors and awards

2000 Bayer Corporation Award for Excellence in Chemistry, 2000
2002 College of Science and Mathematics Dissertation Fellowship
2004 Lineberger Comprehensive Center NIH NSRA Postdoctoral Fellowship
2013 American Crystallographic Association Margaret C. Etter Early Career Award
2013 Albert E. Levy Scientific Research Award for Junior Faculty
2013 Department of Biochemistry, Emory University Outstanding Teaching Award
2015 Awarded the W. M. Keck Foundation Medical Research Grant
2017 Postdoctoral Office “One in a hundred” Mentor of the Year Award
2017 “Special Recognition for Outstanding Research” Emory University School of Medicine

C. Contributions to Science

C. Contributions to Science

Nuclear Receptor Structure-Function
Our lab has taken a multi-faceted approach to understand how NRs are activated by small molecules. We have extensively studied the glucocorticoid receptor to shed light on ligand recognition and discrimination. This include studies of selective GR agonists as well as ligands currently in clinical trials. We have also generated the first small-molecule bound structures of the liver receptor homolog 1 (NR5A2, LRH-1) and have a robust collaboration at Emory to design and synthesize potent and selective agonists for this receptor.

a) Flynn, A.R., Mays, S.G., **Ortlund, E.A.**, Jui, N.T., Development of Hybrid Phospholipid Mimics as Effective Agonists for Liver Receptor Homolog-1, *ACS Med Chem Lett*., 2018, 9(10):1051-1056. PMID PMC6187417
b) Mays, S.G., Okafor, C.D., Whitby, R.J., Goswami, D., Stec, J., Flynn, A.R., Dugan, M.C., Jui, N.T., Griffin, P.R., **Ortlund, E.A**., Crystal Structures of the Nuclear Receptor, Liver Receptor Homolog 1, Bound to Synthetic Agonists, *Journal of Biological Chemistry,* 2016. 291:49 PMCID PMC5207232
c) Mays S., Okafor D.C, Whitby R.J., Dharmarajan V., Stec J., Griffin, P.R., **Ortlund E.A.**, Structure and Dynamics of the Liver Receptor Homolog 1-PGC1α Complex. *Molecular Pharmacology,* 2017, 92(1):1-11. PMCID PMC5452058
d) Kohn JA, Deshpande K, **Ortlund EA**. Deciphering modern glucocorticoid cross-pharmacology using ancestral corticosteroid receptors. *Journal of Biological Chemistry*. 2012:287:16267-75 PMCID: PMC3351298.

2. Steroid Receptor Evolution

Combining phylogenetics with structural biology and biochemistry, we have made seminal impacts in understanding how tight molecular partnership evolve. Currently, we are interested in 1) how proteins evolve new functions (not simply shifts ligand specificity or changes in stability), 2) how intergenic noncoding RNAs acquire function and 3) how proteins evolve the ability to perform multiple functions. This is an extremely intellectually challenging and fun area of science that is making a big impact in how we understand the forces that shaped modern biology. While the tools to achieve this have been around for decades, their use has largely been contained to their respective biological fields. Ultimately, our goal is to propel the broad adoption of a combined phylogenic/ biochemical approach in all areas of biology. We have largely focused on steroid receptors which are an ideal system studying the evolution of molecular recognition and allostery.

a. **Ortlund EA**, Bridgham JT, Redinbo MR, Thornton JW. Crystal Structure of an Ancient Protein: Evolution by Conformational Epistasis. Science. 2007:317:1544-8. PMCID: PMC2519897
b. Bridgham JT, **Ortlund EA**, Thornton JW. An Epistatic Ratchet Constrains the Direction of Glucocorticoid Receptor Evolution. Nature. 2009:461:515-9. PMID: 19779450.
c. Harms M.J., Eick G.N., Goswami D., Colucci J.K., Griffin P.R., **Ortlund EA**, Thornton J.W. Biophysical mechanisms for large-effect mutations in the evolution of steroid hormone receptors. Proceeding of the National Academy of Sciences U.S.A. 2013:110:11475-80. PMCID: PMC3710831.
d. Hudson WH, Kossmann, BR, S. de Vera, IM, Chuo, S, Weikum, ER, Eick, G, Thornton, JW, Ivanov, I, Kojetin, DK, **Ortlund EA**. Distal substitutions drive divergent DNA specificity among paralogous transcription factors through a subdivision of conformational space, Proceedings of the National Academy of Sciences U S A, 113(2):326-31. PMCID: PMC4720311.

3. Phospholipid-regulated gene expression

During my postdoctoral training, I tackled the characterization of liver receptor homologue -1 (LRH-1), an orphan nuclear receptor involved in glucose homeostasis, reverse cholesterol transport, lipid absorption, hormone synthesis, and cell proliferation. My structural and functional studies identified phospholipids as *bona fide* LRH-1 ligands, identifying LRH-1 as the first mammalian phospholipid-sending transcription factor. We have continued to pursue this challenging project attempting to link phospholipid sensing with glucose, lipid, and bile acid homeostasis. We have recently established a collaboration with Dr. David Cohen (Harvard Medical School/BWI) centered on the action of START domain containing proteins and their connection to phospholipid-driven gene regulation. Together, we have discovered that PC-TP stimulates LRH-1 transcriptional activity making PC-TP a critical player in the phosphatidylcholine signaling pathway that impacts glucose homeostasis. We are interested in the molecular mechanisms that drive the unique biology of phospholipids to control metabolism.

a. **Ortlund EA**, Lee Y, Solomon IH, Hager JM, Safi R, Choi Y, Guan Z, Tripathy A, Raetz CR, McDonnell DP, Moore DD, Redinbo MR. Modulation of human nuclear receptor LRH-1 activity by phospholipids and SHP. Nat Struct Mol Biol. 2005:12:357-63. PMID: 15723037
b. Lee JM, Lee YK, Mamrosh JL, Busby SA, Griffin PR, Pathak MC, **Ortlund EA**, Moore DD. A nuclear-receptor-dependent phosphatidylcholine pathway with antidiabetic effects. Nature. 2011:474:506-10 PMCID: PMC3150801.
c. Musille PM, Pathak MC, Lauer JL, Hudson WH, Griffin PR, **Ortlund EA**. Antidiabetic phospholipid-nuclear receptor complex reveals the mechanism for phospholipid-driven gene regulation. Nat Struct Mol Biol. 2012:19:532-7. PMCID: PMC3960984.
d. Musille PM, Pathak M, Lauer JL, Griffin PR, **Ortlund EA**. Divergent sequence tunes ligand sensitivity in phospholipid-regulated hormone receptors. J Biol Chem. 2013:288:20702-12 PMCID: PMC3711333.

4. Phospholipid transport and signaling

Since 2006, I have studied how phospholipids are sensed and signal though interaction with the Sec14 family of proteins. This has largely been a collaborative effort between our lab and Dr. Vytas Bankaitis (Texas A&M Health Science Center). I was responsible for discovering the molecular mechanism driving both phosphatidylcholine and phosphatidylinositol recognition in the protein family. We were able to develop mutant variants with selective disruptions in their ability to transport either PC or PI and tie specific

lipid shutting abilities to cell functions such as proliferation, vesicle maturation, and protein synthesis. We have extensive experience in manipulating the phospholipid transfer ability of most known soluble phospholipid transport proteins in both yeast and mammals.

a. Schaaf G, **Ortlund EA**, Tyeryar KR, Mousley CJ, Ile KE, Garrett TA, Ren J, Woolls MJ, Raetz CR, Redinbo MR, Bankaitis VA. Functional anatomy of phospholipid binding and regulation of phosphoinositide homeostasis by proteins of the sec14 superfamily. Mol Cell. 2008:29:191-206. PMID: 18243114.
b. Schaaf G, Dynowski M, Mousley CJ, Shah SD, Yuan P, Winklbauer EM, de Campos MK, Trettin K, Quinones MC, Smirnova TI, Yanagisawa LL, **Ortlund EA**, Bankaitis VA. Resurrection of a Functional Phosphatidylinositol Transfer Protein from a Pseudo-Sec14 Scaffold by Directed Evolution. Mol Biol Cell. 2011:22:892-905. PMCID: PMC3057712.
c. Ren J, Pathak MC, Temple BRS, Lin C, Nile AH, Mousley CJ, Duncan MC, Eckert DM, Leiker TJ, Ivanova PT, Meyers MS, Murphy RC, Brown HA, Verdaasdonk J, Bloom KS, **Ortlund EA**, Neiman AM, Bankaitis VA A Phosphatidylinositol Transfer Protein Integrates Phosphoinositide Signaling With Lipid Droplet Metabolism To Regulate a Developmental Program of Nutrient Stress-Induced Membrane Biogenesis. Molecular and Cellular Biology. 2014:25(5): 712-727. PMCID: PMC3937096.
d. Tillman, M.C., Khadka, M., Duffy, D., Wang, M.C., **Ortlund, E.A.**, Structural characterization of life-extending *Caenorhabditis Elegans* Lipid Binding Protein 8, *Science Reports*, 2019, 9, Article number: 9966. PMCID: PMC6620326.

Complete List of Published Work in MyBibliography (64 total):

https://www.ncbi.nlm.nih.gov/myncbi/eric.ortlund.1/bibliography/public/

D. Additional Information: Research Support

Ongoing Research Support

Winship Invest$ Pilot Grant Ortlund (PI) 06/01/2019 – 05/31/2020 Emory
CRISPR/Cas-based High-Throughput Protein-RNA Interaction Screen to Identify lncRNAs Functionally Associated with Transcription Factors in Cancer

R01 DK56626 Cohen (PI) 04/01/2015 - 03/31/2020 NIH/NIDDK
Phospholipid-Mediated Metabolic Control in Liver and Brown Adipose Tissue
To explore mechanisms by which membrane phosphatidylcholines regulate metabolism in liver and brown fat.

R01 DK103046 Cohen (PI); Ortlund (mPI) 07/01/2015 - 06/30/2020 NIH
Them1-Mediated Metabolic Regulation and Pathogenic Role in NAFLD
To elucidate Them1-mediated metabolic regulation and identify new therapeutic targets to manage NAFLD.

Medical Research Grant Ortlund (PI) 03/01/2015 - 02/28/2020 Keck
W. M. Keck Foundation
Deciphering the Evolution of Molecular Recognition and Gene Regulation Using Resurrected Molecules.
The goal of this proposal is to determine now GR evolved the ability to recognize new DNA sequences to directly suppress gene expression.

1R01DK115213-01 Ortlund (PI); Calvert and Jui (mPI) 07/01/2017 - 06/30/2022 NIH
Targeting LRH-1 with Small Molecule Modulators
The goal of this project is to develop and characterize synthetic small molecule modulators of LRH-1.

R01MH082833 Maney, (PI) 04/01/2017 – 03/31/2022 NIH/NIMH
A Unique Natural Model for Studying the Mechanisms Underlying Social Behavior
The goal is to use a natural wild model system with defined genotype-phenotype associations to test for causal connections between gene variants and social behavior, at multiple levels of biological organization.

1627789 Maney, PI; Ortlund coPI 07/01/2017 – 06/30/2020 NSF
A Model of Behavioral Evolution from Genotype to Phenotype

To develop a vertically integrated model of how genetic change leads to phenotypic change wild vertebrates.

HHSN272201300018I/HHSN27200009 Rouphael/ Anderson (PI) 09/01/2015 – 01/03/2023 NIH
Vaccine Treatments Evaluation Unit (VTEU); Consultation and 'omics testing of clinical samples
Our obligation is to perform lipidomics on 400-1200 clinical samples per year with the goal of identifying lipid pathways and biomarkers for vaccine efficacy against Tullaremia and Yellow Fever. **Lipidomics Core Facility, No funding for the Ortlund lab.*

1U24DK112341-01 (MPI) Ortlund (contact mPI); Fernandez (mPI) 09/01/2016 – 08/31/2022 NIH
Georgia Comprehensive Metabolomics and Proteomics Unit for MoTrPAC
This grant is in response to in response to RFA-RM-15-011 "Molecular Transducers of Physical Activity Metabolomics and Proteomics Chemical Analysis Sites" (U24). **Lipidomics Core Facility, No funding for the Ortlund lab.*

1R01AG057470 Hajjar (contact mPI), Seyfried (mPI) ; 09/01/2017 – 08/31/2020 NIH/NIA
Building a high-resolution multi-omic AD interactome with the AMP-AD and M2OVE-AD Projects
The goal of this proposal is to develop a novel plasma protein biomarker platform for AD. **Lipidomics Core Facility, no funding for the Ortlund Lab.*

<u>COMPLETED</u> in the last 3 years

R01 NS037112 NIH/NS Hepler (PI)	06/01/2014 – 05/30/2017
Internal Award Emory Pediatrics Emory Jones (PI); Ortlund (PI)	07/01/2016 – 06/30/2017
R01 DK095750 NIH/NIDDK Ortlund (PI)	05/01/2012 – 04/30/2018
R01 GM090158 NIH/NIGMS Kahn (PI)	07/01/2015 – 05/31/2020
Barth Syndrome Foundation Idea Research Grant Ortlund (PI)	03/01/2016 – 02/28/2019
272201300018I-0-27200006-1 NIH/NIAID Rouphael/ Anderson (PI)	07/25/2015 – 10/31/2019

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
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NAME: Patel, Anamika

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

POSITION TITLE: Assistant Professor (Research Track), Department of Biochemistry

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)*	Completion Date MM/YYYY	FIELD OF STUDY
Mohanlal Sukhadia University, India.	B.S.	07/1997	Chemistry, Botany, Zoology
Mohanlal Sukhadia University, India.	M.S.	07/1999	Chemistry
Central Salt & Marine Chemicals Research Institute, India	Ph.D.	11/2004	Chemistry
Technical University, Munich, Germany	Postdoctoral	10/2005	Biochemistry
Syracuse University, Syracuse, NY.	Postdoctoral	12/2009	Biochemistry

A.Personal Statement

I began my scientific training in the field of biochemistry and structural biology in 2001 and was intrigued by how you could visualize 3-D structures of proteins and link form to function. Since then, I've remained interested in studying the structure-function relationship of proteins, exclusively focusing on the field of transcriptional regulation. I possess specific training and expertise in X-ray crystallography, SAXS, cryo-EM, AUC, enzyme kinetics, and various biochemical and biophysical techniques required to study protein-protein and protein-DNA interactions relevant to this proposal. I started my postdoctoral training in Dr. Michael Cosgrove's laboratory at Syracuse University, where I was exposed to the interesting functions of chromatin modifying enzymes in transcriptional regulation and their significance in numerous diseases. During my stay in his lab, I worked on research projects that involved understanding the molecular mechanism of multiple histone H3 lysine 4 (H3K4) methylation catalyzed by the Mixed Lineage Leukemia-1 (MLL1) Core Complex, with an ultimate goal to provide a framework to develop a new class of therapies for MLL1-related leukemias. My work in his lab answered some long-standing questions in the field such as: how this five-protein complex gets assembled, and how the H3K4 tri-methylation activity of MLL1 is regulated by its complex components. These results led to a paradigm-shift in the field and suggested a novel mechanism used by this family of enzymes. I was also able to determine the structure of MLL1 in complex with one of its interacting proteins providing the basis for rational design of new class of drugs to target hyperactive form of MLL1 in some cases of leukemia.

After joining Department of Biochemistry at Emory University as an Assistant Professor (Research track), I have continued my research in the area of transcriptional regulation. I worked on a highly diverse class of transcription factors containing a tandem array of C2H2-type Zinc fingers in the group of Dr. Xiaodong Cheng at Emory. I solved several structures by X-ray crystallography and published few high-profile papers explaining how Zinc-fingers deviate from their conventional code of DNA recognition to achieve high sequence specificity without any gross penalties on DNA-binding. Recently, I shifted my focus towards cryo-EM due to two main reasons: first, my persistent interest in visualizing large transcriptional complexes and second, the technological advancement in cryo-EM now allow high-resolution structure determination. Driven by these interests, I undertook a research project in Dr. Eric Ortlund's lab at Emory to study the mechanism of transcriptional regulation by the liver receptor homologe-1, LRH-1. LRH-1 is a member of the nuclear receptor family of transcription factor and it is activity regulated at multiple levels by ligand binding, DNA-binding, and coregulator recruitment for complete transcriptional control. Therefore, determining the structure of LRH-1 in complex with DNA and co-regulator proteins makes perfect sense. Given the fact that the complex is resistant to crystallization, cryo-EM is the only available tool to derive structural information. I produced good preliminary

results after some rounds of optimization that have laid a solid groundwork for current proposal. Over the past few years, I have attended a few workshops and training sessions to refine my skills in cryo-EM; these points, in addition to the strong collaborative support we have, make us fully capable to carry out the proposed research.

B.Positions and Honors
Positions and Employment

1999-2001: Project Assistant at Central Salt & Marine Chemicals Research Institute, Bhavnagar, India.
2006-2009: Postdoctoral Research Fellow at Department of Biology, Syracuse University, Syracuse, NY.
2010-2011: Assistant Professor (Research Track), Department of Biology, Syracuse University, Syracuse, NY
2012-2013: Assistant Professor (Research Track), Department of Biochemistry and Molecular Biology, SUNY Upstate Medical University, Syracuse, NY
2013-present: Assistant Professor (Research Track), Department of Biochemistry, Emory University School of Medicine, Atlanta

Other Experience and Professional Memberships

Member of American Association of Advancement of Science (AAAS)
Member of American Association of Cancer Research (AACR)

Honors

2005: DAAD Short Term Research Fellow at Physics department, Technical University, Munich, Germany.

1. C. Contributions to Science

Zinc Finger proteins and sequence specific DNA recognition.

The C2H2 type zinc finger (ZF) proteins belong to the largest and most diverse family of transcription factors in human. They have well documented functional role in DNA binding. Despite being the largest family, the molecular targets and the biological function for the majority of them remains elusive. Based on structural knowledge of 2-3 tandem ZF, algorithms are derived to predict their DNA binding sequences. Yet, often they do not match with experimentally identified sequences, specifically in case of ZF array containing large number of tandem fingers. This suggesting the need for the structural studies of a tandem ZFs-DNA complex to enhance our understanding in their code of DNA recognition. My structure-function studies on two of these ZF proteins PRDM9 and ZFP568, each containing 13 and 11 tandem ZF, respectively, help to decipher the molecular basis of their DNA recognition. PRDM9 that directs the sequence specific binding to meiotic recombination hotspots and showed adaptability in their ZFs by flipping two differently positioned residue to the variations in DNA sequence and thus, allows them to respond to the variation in DNA sequences without any gross penalties in their binding affinities. My studies help to explain genetic data on how allelic variants of PRDM9 could influence their DNA binding targets and therefore, their meiotic recombination landscape. Along this line, I worked on a project in collaboration with Dr. Todd Macfarlan at NIH on a ZF protein ZFP568 that directly regulates the expression of a placental growth factor Igf2-P0 and is required for embryonic development in mice. I determined the structure of ZFP568 in complex with DNA, this is first structure available to date showing the largest number of ZFs in complex with DNA. The structures, I determined are of significant importance as they show deviations at several levels from the classical C2H2 type ZF DNA recognition code. The structures propose flexibility in the ZF by 2-,3-or 4-base pair specific recognition per zinc finger to enhance the binding capacity and also respond to the specific DNA shape. Collectively, these structures provide additional layers of adaptable binding ability between ZFs and DNA.

a. **Anamika Patel**, Peng Yang, Matthew Tinkham, Mihika Pradhan, Ming-An Sun, Yixuan Wang, Don Hoang, Gernot Wolf, John R. Horton, Xing Zhang, Todd Macfarlan and Xiaodong Cheng. DNA Conformation Induces Adaptable Binding by Tandem Zinc Finger Proteins (2018). *Cell*, 147(1) 221-233. PMCID:PMC5877318

b. **Anamika Patel,** Xing Zhang, Robert Blumenthal and Xiaodong Cheng. Structural basis of human PR/SET domain 9 (PRDM9) allele C-specific recognition of its cognate DNA sequence (2017). *Journal of Biological Chemistry*, 292(39) 15994-16002.PMCID:PMC5625032

c. **Anamika Patel**, J.R. Horton, G.G. Wilson, X. Zhang, X. Cheng. Structural basis for human PRDM9 action at recombination hot spots (2016). *Genes & Development*. 2016. 30: 257-265. *(This work was featured on the journal's cover.)* PMCID:PMC4743056

d. **Anamika Patel**, H. Hashimoto, Xing Zhang, Xiaodong Cheng. Characterization of How DNA Modifications Affect DNA Binding by C2H2 Zinc Finger Proteins (2016). *Methods in Enzymology, 573 387-403*

2. MLL1-WDR5-RbBP5-Ash2L-DPY30: a core complex responsible for histone H3 lysine 4 tri-methylation

The disruption of Mixed Lineage leukemia-1 (MLL1) is frequently observed in acute lymphoblastic or acute mylogenous leukemia, which results in either increased or decreased MLL1's function. The MLL1 protein catalyzes histone H3 lysine 4 tri-methylation and regulates chromatin structure for active gene expression. The catalytic activity of MLL1 is tightly regulated by its core components. One question of significant importance in the field is that how MLL1's activity is regulated by its core components. My work showed that unlike other methyltransferase MLL1 core complex uses a novel mechanism involving two active sites to regulate its activity. We have identified a interaction motif in MLL1, which we named as "WIN" motif (also present in other family members: MLL2/3/4, SET1A and SET1B) required for binding with one of its regulatory subunit WDR5 and solved the first structure of MLL1 WIN motif in complex with WDR5. We also showed that targeting the interaction surface of MLL1-WDR5 successfully inhibits MLL1's activity and thus, provide a novel approach to target MLL1's activity in leukemia. As evidence of the high impact of this work, this research was published in several peer-reviewed journal articles as mentioned below. One article was pushed for accelerated publication and was featured on the cover of the Journal of Biological Chemistry. A second paper was highlighted as the paper of the week in the same journal.

a. **Anamika. Patel**, V.E. Vought, V. Dharmarajan and M.S. Cosgrove (2011) A novel non-SET domain multi-subunit methyltransferase required for sequential nucleosomal histone H3 methylation by the MLL1 core complex. *Journal of Biological Chemistry* **286**(5): 3359-3369. PMCID: PMC3030342

b. **Anamika. Patel,** V. Dharmarajan, V.E. Vought and M.S. Cosgrove (2009) On the mechanism of multiple lysine methylation by the human Mixed Lineage Leukemia Protein-1 (MLL1) core complex. *Journal of Biological Chemistry* **284**(36): 2424-24256. PMCID: PMC2782018 *(**selected as the paper of the week and spotlight the first author**).*

c. **Anamika. Patel,** V. Dharmarajan and M.S. Cosgrove (2008) Structure of WDR5 bound to Mixed Lineage Leukemia Protein-1 peptide. *Journal of Biological Chemistry* **283**(47): 32158-32161. PMID: 18829459. (***Accelerated Publication*, *Featured on the Journal Cover***).

d. **Anamika. Patel,** V.E. Vought, V. Dharmarajan and M.S. Cosgrove (2008) A conserved arginine containing motif crucial for the assembly and enzymatic activity of the Mixed Lineage Leukemia protein-1 core complex. *Journal of Biological Chemistry* **283**(47): 32162-32175. PMID: 18829457. (***Featured on the Journal Cover***).

3. Analytical Ultracentrifugation (AUC) to study protein-protein interaction and conformational switch

I have my expertise in Analytical Ultracentrifugation to study Protein-Protein interactions and conformational changes in proteins in collaboration with various groups.

a. S. Zhong, F. Hsu, C.J. Stefan, X. Wu, **Anamika. Patel**, M.S. Cosgrove and Y. Mao (2012) Allosteric activation of the phosphoinositide phosphatase sac1 by anionic phospholipids. *Biochemistry* **51**(15): 3170-7. PMCID: PMC3329130

b. M. Mbantenkhu, X. Wang, S. Wilkens, E. Hoffman, **Anamika. Patel**, M.S. Cosgrove and X.J. Chen (2011) Mgm101 - a DNA recombinase essential for the mitochondrial DNA maintenance. *Journal of Biological Chemistry* **286**(49): 42360-70. PMCID: PMC3234957.

c. T. Wollert, **Anamika. Patel**, Y.-L. Lee, V.E. Vought, M.S. Cosgrove, J.A. Mercer and G.M. Langford (2011) Myosin5a tail associates directly with Rab3A-containing compartments in neurons. *Journal of Biological Chemistry* **286**(16): 14352-14361. PMCID: PMC3077635.

4. Bili-proteins in cyanobacteria as photo-optical switch

My earlier work showed biochemical and structural characterization of photosynthetic bili-proteins in cyanobacteria from marine and fresh water environment. I showed that bili-proteins isolated from marine cyanobacterial species have higher thermal stability and antioxidant potential compared to fresh water species. We were able to determine for the first time a structure of a photo-optical switch of so-called E-isomer in bili-proteins.

a. M. Schmidt, **Anamika. Patel**, Y. Zhao and W. Reuter (2007) Structural Basis for the Photochemistry of R-Phycoerythrocyanin. *Biochemistry* **46**: 416-423. PMID: 17209552

b. L. Satyanarayana, C.G. Suresh, **Anamika. Patel**, S. Mishra and P.K. Ghosh (2005) X-ray crystallographic studies on C-phycocyanin from the cyanobacteria of different habitats: marine & fresh water. *Acta Crystallographica Section F: Structural Biology and crystallization communications* F61: 844-847. PMCID: PMC1978106.

c. B.T. Paul, **Anamika. Patel**, G.S. Selvam, S. Mishra, P.K. Ghosh and R. Murugesan (2006) Photodynamic action of C-phycocyanins obtained from marine and fresh water cyanobacterial cultures: A comparative study using EPR spin trapping technique. *Free Radical research* **40**: 821-825. PMID: 17015260

d. **Anamika. Patel**, S. Mishra, R. Pawar, S. Sonawane and P. K. Ghosh (2005) Purification and characterization of C-Phycocyanin from cyanobacterial species of marine and fresh water habitats. *Protein Expression and Purification* **40**: 248-255. PMID: 15766866

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

Ongoing

None

Completed

5 R01 GM 049245NIH/NIGMS (Subaward) Cheng (PI), Patel, (co-I) 04/01/2017 -07/31/2017
DNA Methylation: Structures, Functions and Regulation
The goal of this grant was to determine central aspects, enzymatically and structurally, of (1) 5mC oxidation by Tet proteins, (2) modification-specific recognition by C2H2 zinc-finger and SRA-domain proteins, and (3) 5mC and 5hmC base excision by DNA glycosylases.