BIOGRAPHICAL SKETCH

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NAME: Rebecca Page

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

POSITION TITLE: Professor of Cell Biology

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
University of Arizona, Tucson, Arizona	B.S.	05/1993	Biochemistry
University of Arizona, Tucson, Arizona	B.S.	05/1993	Applied Mathematics
Princeton University, Princeton, NJ	Ph.D.	01/2000	Chemistry/X-ray Cryst.
The Scripps Research Institute, La Jolla, CA	Post-doc	03/2003	Molecular Biology, X-ray Crystallography

A. Personal Statement

My group has a long-standing interest in signaling pathways regulating cell proliferation/differentiation and their derangement in human diseases. **A major focus of my work is defining the role of ser/thr phosphoprotein phosphatases (PPPs) in these pathways,** with a long-term goal of elucidating how PPPs dephosphorylate thousands of different protein substrates while allowing the level of phosphorylation to be individually and exquisitely regulated. The goal of this proposal is to elucidate how the PPP holoenzyme **PP6** assembles with, recruits and dephosphorylates its substrates, in order to define PP6 function in inflammation, DNA damage and the cell cycle, among other pathways. To achieve this, my group integrates structural, biophysical, biochemical and cell biological methods to define how PPPs, and their interaction with regulatory proteins and substrates, control phosphorylation signaling in cells. **We have made fundamental contributions to this field,** by, for example: (1) discovering the PP1 regulatory code used by PP1-specific regulators to direct the activity of PP1, (2) identifying the LxVP short linear motif (SLiM) binding site on CN, which revealed that immunosuppressants inhibit CN activity by blocking its ability to bind substrates; (3) the discovery that the B-subunits of PP2A engage regulators and substrates using a phosphorylation-specific SLiM (B56) and, most recently, short helical motifs (SHelMs; B55), among others. These data are transforming our understanding of the highly specific and exquisitely regulated function of PPPs in biological signaling and disease. Here, we will apply our decades of experience to defining the architecture of PP6 holoenzymes and the mechanism(s) by which it binds substrates.

Dr. Peti and I are long-term collaborators, having co-authored scores of papers focused on elucidating the structural and functional basis of PPP regulation in cells and cancer, with additional manuscripts in preparation. I am also a long-time collaborator of Dr. Kettenbach (we have co-authored six manuscripts including work defining PPP substratomes). I have been funded since 2008 by the NIH, NSF (NSF-CAREER award), ACS (Research Scholar) among others. I have published 139 papers, many in high impact journals. My broad background in signaling and biophysics enables me to productively collaborate on the experiments outlined in this proposal, for which structure determination by cryo-EM and crystallography are key components. I have directly mentored dozens of post-doctoral scientists, graduate students and undergraduates throughout my career, with alumni from my group having obtained highly competitive positions (senior crystallographer at Bristol-Myers Squibb, faculty position at Vanderbilt University among others; 2 of my 6 graduate students received awards for the best dissertation either at the school, Brown University, or departmental, MCB Brown University, level). The research environment at University of Connecticut Health Center is exceptional, allowing my group to thrive both academically and scientifically. Clearly, my training, my grant management skills, as well as my research, especially in the fields of phosphatases and signaling, demonstrate that I am highly qualified to contribute to the exciting, transformative studies outlined in this proposal.

References (collaborators on this proposal in bold)

(**A1**) Choy, MS, Moon, TM, Ravindran, R, Bray, JA, Robinson, LC, Archuleta, TL, Shi, W, **Peti, W**, Tatchell, K **Page, R** (2019) SDS22 selectively recognizes and traps metal-deficient inactive PP1. ***PNAS*** .116: 20472-20481. PMID: 31548429

(**A2**) Wang, X., Garvanska, DH, Nasa, I, Ueki, Y, Zhang, G., **Kettenbach, AN**, **Peti, W**, Nilsson, J, **Page, R** (2020) A dynamic charge-charge interaction modulates PP2A: B56 substrate recruitment. ***ELife.*** 9: e55966. PMID 32195664.

(**A3**) Padi, SKR, Vos, MR, Godek, RJ, Fuller, JR, Kruse, T, Hein, JB, Nilsson, J, Kelker, MS, **Page R**, **Peti W**. (2024) Cryo-EM structures of PP2A:B55-FAM122A and PP2A:B55-ARPP19. ***Nature***. 625:195-203. PMID: 38123684.

Ongoing and recently completed projects include:

1R01GM144379
Page (PI)
NIH - NIGMS
05/01/2023 – 04/30/2027
The regulation of PP1 in the nucleus

Project Grant
Page (Collaborator)
Open Philanthropy
01/01/2022 – 12/31/2026
Syphilis Vaccine Development

1R01NS124666
Page (Collaborator)
01/01/2023 – 12/31/2027
Serine/Threonine Phosphatases in Neurological Diseases

NX012-107-160-002
Page (PI)
01/03/2019 – 07/31/2022
Biophysical analysis of Novel Allosteric SHP2 Inhibitors

B. Positions and Honors

Academic Appointments

12/2020 – present	Professor, Department of Cell Biology, School of Medicine, University of Connecticut Health Center, Farmington, CT
12/2020 – present	Professor, Department of Molecular Biophysics and Biochemistry, School of Medicine, University of Connecticut Health Center, Farmington, CT, *secondary appointment*
7/2019 – 6/2020	CBC Associate Department Head of Academic and Faculty Affairs, *Interim*, University of Arizona, Tucson, Arizona
1/2019 – 12/2020	Professor, Department of Immunobiology, University of Arizona, Tucson, Arizona
1/2017 – 12/2020	Professor and Donna B. Cosulich Faculty Chair, Departments of Medicine and Chemistry & Biochemistry, University of Arizona, Tucson, Arizona
7/2015 – 12/2016	Professor of Biology, Molecular Biology, Cell Biology and Biochemistry, Brown University, Providence, RI
7/2011 – 6/2015	Associate Professor of Biology, (Tenured), Molecular Biology, Cell Biology and Biochemistry, Brown University, Providence, RI
7/2005 – 6/2011	Assistant Professor of Biology (Tenure-track), Molecular Biology, Cell Biology and Biochemistry, Brown University, Providence, RI
9/2004 – 6/2005	Assistant Professor of Medicine (Research), Molecular Pharmacology, Physiology and Biotechnology, Brown University, Providence, RI
4/2003 – 8/2004	Core Director, Crystallomics Core, Joint Center for Structural Genomics at The Scripps Research Institute, La Jolla, CA

Honors and Awards

6/2021	Keynote speaker, PDB50 sessions of the 2021 ACS National Meeting
2019	UBRP mentor of the year finalist, University of Arizona
2017 – 2020	Donna B. Cosulich Faculty Fellow, University of Arizona
2016 – 2020	NIH – MSFB, *permanent member*
2013	Eighteenth Annual Gehrenbeck Lecturer, Rhode Island College
2010 – 2015	NSF-CAREER award
2008	Hazeltine Citation Nominee (Brown University senior class teaching award)
2008 – 2011	American Cancer Society (ACS) Research Scholar
2006	Brown University Seed Fund Award
2005	Richard B. Salomon Faculty Research Award

2000 – 2003 Ruth L. Kirschstein National Research Service Award (NRSA), NIH
1997 – 1998 Harold W. Dodds Honorific Graduate Fellowship
1994 – 1997 National Science Foundation Graduate Fellowship
1993 Phi Beta Kappa
1992 – 1993 Barry M. Goldwater Undergraduate Research Scholar
1988 – 1992 Regents Academic Achievement Scholarship

Other Experience

Editorial

1/2017 – present Associate Editor, *Science Advances*
7/2012 – 2017 Editorial Board Member, *Journal of Biological Chemistry*
2005 – 2012 Associate Editor, *Protein Expression and Purification*
2003 – present *Reviewer for:* Nature Chemical Biology, Nature Communications, Nature Methods, Nature Structural & Molecular Biology, Science Signaling, eLife, PLoS Pathogens, Acta Cryst D/F, Biochem, J Biol Chem, FASEB J, J Mol Biol, Mol Cell Bioc, Mol Microbiol, Prot Exp Purif, Proteins: Struc Func Bioinformatics, Protein Science, *among others*

Study Sections/Reviewer

2023-present National Center for CryoEM Access and Training (NCCAT), invited proposal reviewer
2021 – 2023 NSLSii User Executive Committee, elected member
2019 External Scientific Advisory Board member, NSLS II, Brookhaven National Laboratory
2019 – 2022 SSRL User Executive Committee, elected member
07/2016-09/2020 NIH MSFB (Macromolecular Structure and Function), *permanent member*
6/2016 – 5/2019 BNL National Synchrotron Light Source II scientific proposal review panel, *standing member*
7/2015 NIH ZRG1 BCMB-X (40) P01 review panel, *Ad hoc study section member*
7/2015 NIH ZRG1 BCMB-W (40) P41 *Ad hoc reverse site visit study section member*
2/2015 NIH NIGMS P41 pre-proposal (X02) review panel, *Ad hoc study section member*
2014 – 2016 Structural Biology Proposal Study Panel (PSP), ALS, LBNL, *Standing reviewer*
11/2013 NIH – MSFC (Macromolecular Structure and Function), *Ad hoc panel member*
7/2012 NIH – NCRR, ZRG1 BCMB P41 *Ad hoc site visit study section member*
10/2010 NSF – MCB, Cellular Systems Cluster, Cellular Homeostasis and Signaling, Ad Hoc review panel member
10/2010 Temporary Member, NIH, NCI Innovative Molecular Analysis Technologies Program
3/2010 Temporary Member, NIH, NCI Innovative Molecular Analysis Technologies Program
2010 The Welcome Trust, Scientific Grant Reviewer
2009 Cancer Research UK, Project Grant Application Reviewer
2009 NSF Reviewer, Molecular Biochemistry Program
2007 Cancer Research UK, Project Grant Application Reviewer

Meeting Organizer/session chair

2017 Session Chair, Bacterial communities and mechanisms of microbial cell regulation, Session of the 2017 American Society of Biochemistry and Molecular Biology (ASBMB) meeting; Chicago, Il
2016 Co-organizer, Protein Structure Dynamics & Function: Sailing the Protein Seas, Providence, RI
2013 Session Chair, Structural Enzymology (I) Session of the 2013 American Crystallographic Association Meeting.
8/30/2008 Session Chair, Focused Structural Proteomics Microsymposium, XXI congress and General Assembly of the International Union of Crystallography, Osaka, Japan.
2/10/2008 Session Chair, Protein Expression Research Group Session, Association for Biomolecular Resource Facilities Annual Meeting, Salt Lake City, Utah, USA.

C. Contributions to Science: Total publications: 139; PI and collaborators in bold; see https://www.ncbi.nlm.nih.gov/myncbi/rebecca.page.1/bibliography/public/

(i) Mechanism of action and function of the ser/thr phosphoprotein phosphatases PP2A and CN. PP2A is a ser/thr phosphatase that is essential for mitosis and cancer. Different from PP1, it forms trimeric holoenzymes that differ in the nature of the 'B' subunit (substrate specifying subunit). While it was thought for decades that SLiM-based regulator binding was restricted to PP1, we and others showed that CN and PP2A:B56 use PPP-

specific SLiMs to bind their cognate substrates/regulators, strongly suggesting that this SLiM-based mechanism is conserved throughout the entire family. Specifically, we showed that CN binds its substrates using the LxVP SLiM (in addition to the PxIxIT SLiM) and, further, that the LxVP binding site on CN is *exactly where the potent, blockbuster immunosuppressants cyclosporin A and FK506 bind.* We also demonstrated that PP2A:B56 binds specifically to LSPIxE motifs and discovered that this binding is *enhanced* by SLiM phosphorylation (in contrast, phosphorylation of PP1 SLiMs *inhibits* binding), leading to a 'timing mechanism' for PPP activity during mitosis which is regulated by phosphorylation. We also showed that SLiM binding is modulated by dynamic charge-charge interactions, events we expect will be particularly important for other PPP SLiM interactions. Most recently, using high resolution cryo-EM to determine the structures of IDP inhibitors bound to PP2A:B55, we showed that PP2A:B55 binds its inhibitors using short *helical* motifs (SHelMs). In addition to references A1-A3:

(***Ci1***) Grigoriu, S., Bond, R., Cossio, P., Chen, J.A., Ly, N., Hummer, **Page, R.**, Cyert, M.S., **Peti, W.** (2013) The molecular mechanism of substrate engagement and immunosuppressant inhibition of Calcineurin. ***PLoS Biology***, 11(2): e1001492; PMID: 23468591.
(***Ci2***) Hendus-Altenburger, R. Wang, X., Sjogaard-Frich, LM, Pedraz-Cuesta, E. Sheftic, S.R., Bendsoe, A.H., **Page, R.**, Kragelund, B.B., Pedersen, S.F., **Peti, W.** (2019) Molecular basis for the binding and selective dephosphorylation of Nhe1 by CN. ***Nature Communications***, 10: 1.08. PMID: 31375679
(***Ci3)*** Li, Y, Sheftic, SF, Grigoriu, S, Schweiters, CD, **Page, R**, **Peti,, W** (2020). The structure of the RCAN1:CN complex explains the inhibition of and substrate recruitment by calcineurin. ***Science Advances***, 6:eaba3681. PMID: 32936779.
(***Ci4***) Pavic, K., Gupta, N., Domenech, O., Derua, R., Aakula, A., Huhtaniumi, R., Maatta, JA, Hofflin, N., Okkeri, J., Wang, Z., Kauko, O., Roosa, V. Honkanen, H., Abankwa, D., Kohn, M., Hytonen, V.P., Xu, W., Nilsson, J., **Page, R.,** Janssens, V., Leitner, A., Westermark, J. (2023) Structural mechanism for inhibition of PP2A:B56a and oncogenicity by CIP2A. ***Nature Communications***, 14:1143. PMCID:PMC9974998.

(ii) Mechanism of action and function of the ser/thr phosphoprotein phosphatase PP1. PP1, the best-characterized PPP, is regulated by its interaction with more than 200 known *targeting and inhibitor* proteins. This diversity of binding partners is consistent with PP1's regulatory role in multiple cellular processes. Thus, specific PP1 inhibitors or activators hold enormous clinical potential for the treatment of a large variety of diseases. During the last 20 years, we have made substantial contributions to this field that are now leading to paradigm shifts in our understanding of PP1 regulation in the cells. First, our group has determined more structures of *PP1 holoenzymes* than any laboratory in the world. In 2007, only two PP1 holoenzyme structures, of the more than 200 PP1 holoenzymes that exist in the cell, had been determined. This dearth of structural information was due primarily to the exceptional difficulties of studying PP1 in the laboratory. Through a dedicated effort over multiple years, we overcame these challenges. Second, since 2010, we have determined the structures of multiple PP1 holoenzymes as well as additional natural toxin:PP1 complexes. Third, our structures have enabled us to identify completely novel interaction sites leading to the discovery of more than 10 distinct PP1 specific SLiMs. These are key advances for understanding the regulation of PP1. Fourth, we have also advanced the field of protein science in general. This is because most PP1 regulatory proteins are IDPs. This system is unique because it allows us to understand how a family of IDPs bind and interact with a single target, PP1. This is an exciting and growing field as more than 30% of all human proteins are predicted to have large IDP domains. Here, we are leveraging these discoveries to develop novel tools—PhosTAPs and PhosTACs—to identify all residues directly regulated by PP1 and to manipulate target phosphorylation levels, respectively.

(***Cii1***) Choy, M.S., Swingle, M., D'Arcy, B., Abney, K., Rusin, S.F., **Kettenbach, A.N**., **Page, R.**, Honkanen, R.E., **Peti, W**. (2017) PP1:Tautomycetin Complex Reveals a Path toward the Development of PP1-Specific Inhibitors. ***JACS***, 139: 17703-17706. PMID: 29156132
(***Cii2***) Kumar, GS, Choy, MS, Koveal, DM, Lorinsky, MK, Lyons, SP, **Kettenbach, AN**, **Page, R**, **Peti, W**. (2018) Identification of the substrate recruitment mechanism of the muscle glycogen protein phosphatase 1 holoenzyme. ***Science Advances***, 4: 11. 11. PMID: 30443599
(***Cii3***) Srivistava, G., Bajaj, R., Kumar, G.S., Gaudreu-Lapierre, A., Nicolas, H., Chamousset, D., Kreitler, D., **Peti, W.**, Trinkle-Mulcahy, L., **Page, R.** (2022) The ribosomal RNA processing 1B:protein phosphatase 1 holoenzyme reveals non-canonical PP1 interaction motifs. ***Cell Reports***. 41:9:11726. PMID 36450254.
(***Cii4***) Srivistava, G., Choy, M.S., Bolik-Coulon, N., **Page, R**., **Peti, W**. (2023). I3 inhibits Protein Phosphatase 1 via a metal binding dynamic protein-protein interaction. ***Nature Communications***. 14(1):1798. PMID 37002212.

(iii) The regulation of tyr phosphatases by regulatory proteins. The tyrosine phosphatases (HePTP, PTP1B) function to transduce environmental and developmental signals (growth factors, stress) into adaptive and

programmed responses (differentiation, inflammation, apoptosis). We have used an integrated biophysical approach—employing NMR spectroscopy, X-ray crystallography and SAXS—in order to understand how these key signaling PTPs are regulated by their multiple interacting partners. Our work has led to the first structure of the MAPK:PTP complex (p38:HePTP), the structures of an activate and resting state MAPK:PTP complex (ppERK2:HePTP and ERK2:HePTP), and the discovery of new HePTP inhibitors. This work was funded by an American Cancer Society Research Scholar award (RSG-08-067-01-LIB) and is described in >20 publications. Additionally, we have discovered how PTP1B activity is regulated by dynamics and conformation.

(***Ciii1***) Choy, M.S., Li, Y., Machado, L.E.S.F., Kunze, M.B.A., Connors, C., Wei, X., Lindorff-Larsen. K., **Page, R.**, **Peti, W.** (2017) Conformational Rigidity and Protein Dynamics at Distinct Timescales Regulate PTP1B Activity and Allostery. ***Molecular Cell*** 65: 644-658. PMCID: PMC5325675.
(***Ciii2***) Kumar, GS, Clarkson, MW, Kunze, MBA, Granata, D, Wand, AJ, Lindorff-Larsen, K, **Page, R**, **Peti, W.** (2019) Dynamic activation and regulation of the mitogen-activated protein kinase p38. ***PNAS***. 115: 4655-4660. PMCID: PMC5939092.
(***Ciii3***) Torgeson, KR, Clarkson, MW, Kumar, GS, **Page, R**, **Peti, W.** (2020) Cooperative dynamics across distinct structural elements regulate PTP1B activity. ***J. Biol. Chem.*** 295: 13829-13837. PMCID: PMC7535920.
(***Ciii***4) Torgeson, K.R., Clarkson, MW, Granata, D., Lindorff-Larsen, K., **Page, R**., **Peti, W.** (2022) Conserved conformational dynamics determine enzyme activity. ***Science Advances*****.** 8:31, PMCID: PMC9348788.

(iv) Antibiotic resistance and antibiotics/vaccine development. Toxin:antitoxin (TA) systems are two component systems that encode a stable protein 'toxin' whose activity leads to growth arrest and an unstable protein 'antitoxin' or 'antidote' that binds the toxin and mitigates its toxicity. However, persistence and the role of TA systems in initiating the persister state, is one of the most poorly understood mechanisms used by bacteria to survive environmental stress. My group showed that MqsRA defines an entirely novel family of Type II TA systems (communicated in >15 publications). Since 2017, we have extended this work into the study of the target of β-lactams, PBPs. In addition to combining NMR with X-ray crystallography to define the mechanism of resistance in PBP5, we are most excited about our recent fragment screen of PBP4, which has provided a clear roadmap for the development of new, potent enterococci antibiotics (described in this proposal). Finally, we are also using protein engineering coupled with structural biology to develop novel, effective vaccines for syphilis.

(***Civ1***) Wang, X., Kim, Y., Hong, S.H., Ma, Q., Brown, B.L., Pu, M., Tarone, A.M., Benedik, M.J., **Peti, W.**, **Page, R**., and Wood, T.K. (2011). Antitoxin MqsA helps mediate the bacterial general stress response, ***Nature Chemical Biology***, 7, 359-366; PMCID: PMC3097263.
(***Civ2***) Wang, X., Lord, D.M., Cheng, H-Y, Osbourne, D.O., Hong, S.H., Sanchez-Torres, V., Quiroga, C., Zheng, K., Hermann, T., **Peti, W.**, Benedik, M.J., **Page, R**.,* Wood, T.K.* (2012) A New Type V toxin-antitoxin system where mRNA for toxin GhoT is cleaved by antitoxin GhoS, ***Nature Chemical Biology***, 8, 855-861; PMCID: PMC3514572. *co-corresponding authors.
(***Civ3***) **Page, R**., **Peti, W.** (2016) Toxin:antitoxins systems in bacterial growth arrest and persistence. ***Nature Chemical Biology***, 12: 208-214. PMID: 26991085.
(***Civ4***) Hunashal Y, Kumar GS, Choy MS, D'Andrea, E.D., Santiago, A.D.S., Schoenle, M.V., Desbonnet, C., Arthur, M., Rice, L.B., **Page, R., Peti, W.** (2023) The Molecular Basis for Resistance of the ESKAPE bacterium E. faecium Penicillin Binding Protein PBP5 to β-lactam Antibiotics. ***Nature Communications***, 14: 4268. PMID: 37460557.

(v) Advances in structural biology. I made key discoveries to advance structural biology methods.
(***Cv1***) **Page, R**., Grzechnik, S. K., Canaves, J. M., Spraggon, G., Kreusch, A., Kuhn, P., Stevens, R. C. & Lesley, S. A. (2003). Shotgun crystallization strategy for structural genomics: an optimized two-tiered crystallization screen against the *Thermotoga maritima* proteome. ***Acta Crystallogr D*** 59, 1028-1037. PMID: 12777766
(***Cv2***) Canaves, J., **Page, R**., Wilson, I. A. & Stevens, R. C. (2004) Protein biophysical properties that correlate with crystallization success in *Thermotoga maritima*: maximum clustering strategy for structural genomics. ***J Mol Biol***, 344, 977-991. PMID: 15544807
(***Cv3***) **Page, R.***, **Peti, W.***, Wilson, I. A., Stevens, R. C., & Wüthrich, K. (2005). NMR screening and crystal quality of bacterial expressed prokaryotic and eukaryotic proteins in a structural genomics pipeline. ***PNAS***, 102, 1901-1905. *equal contributions; PMCID: PMC548552.
(***Cv4***) Collins, B., Stevens, R.C., **Page, R.** (2005) High-Throughput Optimum Solubility Screening: Using Crystallization Results to Identify the Optimal Buffer for Protein Crystal Formation. ***Acta Crystallogr F****,* 61, 1035-1038; PMCID:PMC1978149.

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: Sathish Kumar Reddy Padi

eRA COMMONS USER NAME (credentials, e.g., agency login): SATISHPADI

POSITION TITLE: Assistant Professor (In Residence), Basic Sciences

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 College of Pharmaceutical Sciences, KU, INDIA	Bachelors in Pharmacy	02/2004	Pharmaceutical Sciences
University of Detroit Mercy, Detroit, MI	MS	08/2008	Chemistry
North Dakota State University, Fargo, ND	PhD	09/2013	Cancer Biology
Medical University of South Carolina, Hollings Cancer Center, Charleston, SC	Postdoc	09/2014	Cancer Biology
University of Arizona Cancer Center, Tucson, AZ	Postdoc	04/2019	Cancer Biology

A. Personal Statement

My long-term research goals involve becoming an independent researcher and understanding comprehensive signaling pathways & mechanisms contributing to drug resistance in various cancers. To pursue these, I plan to leverage my extensive training in diverse fields of cancer pharmacology, structural biology, epigenetics, cell and molecular biology.

As a graduate student in Dr. Bin Guo's laboratory, I identified miR-627 and JMJD1A- as potential targets to exploit the antitumor activity of Vitamin D without eliciting its hypercalcemic side effect. This research was published in the high impact journal Gastroenterology. Due to my outstanding contributions to graduate research, I received numerous awards during my PhD, the Young Investigator Award from SEBM, best graduate student award, among others. As a post-doc in Dr. Andrew S Kraft's laboratory, I addressed the role of PIM kinases on the growth of hematological and solid tumors and elucidating its role in conferring resistance to drug treatment in T-cell Leukemia, prostate, and breast cancers by utilizing PDXs, mouse models, cancer cell lines, organoids and novel PIM inhibitors. We identified, for the first time, that the PIM kinases phosphorylate novel substrates like IRS1, DEPDC5, and EDC3 in various cancer cell lines. I spearheaded projects that unraveled many novel mechanisms underlying drug resistance in various cancers. I was awarded the ASH travel award to present my research. In a different project, I studied the mechanism of DEPDC5 phosphorylation by PIM kinases, a novel mTORC1 regulatory mechanism that establishes PIM as an upstream effector of mTORC1 to promote tumor growth. This study is significant in the context that it reveals a previously unreported mechanistic link between mTORC1 and PIM kinase. For this research, I received the Scholar in training award, one of the most reputed awards for young career scientist presented by the AACR. The research was published in PNAS. In my third project, a collaboration study with Wolfgang Peti's laboratory, we demonstrated that PIM kinases binds to the P-body protein, Enhancer-of-mRNA-decapping-3 (EDC3), and phosphorylates EDC3 on serine (S)161 and this phosphorylation regulates multiple cancer-relevant functions. This research is published in EMBO Reports.

To gain knowledge and further understand the molecular insights into the substrate specificity of Ser/Thr Kinases and Protein Phosphatases, I joined the laboratory of Drs. Wolfgang Peti and Rebecca Page as an Assistant Professor (In Residence)/Basic Sciences at UConn Health, Molecular Biology and Biophysics

Department. Their labs provide an exciting environment for my scientific growth. Frequent interactions with scientists and clinicians through meetings and seminars, aids in awareness about novel approaches in understanding and treating various cancers. My current goal is to achieve an in-depth understanding of signaling networks with a special focus on Ser/Thr kinase and phosphatase using biochemical, biophysical, and structural biology (NMR and Cryo-EM) techniques. Recently we published high resolution Cryo-EM structures of two PP2A:B55-Inhibitor complexes in Nature journal, that highlights how various substrates/inhibitors are recruited to PP2A:B55 and provide a molecular roadmap for the development of therapeutic interventions to target PP2A:B55 related diseases.

Current structural studies in the laboratory are focused on PP6 holoenzyme, a type 2A phosphatase akin to PP2A. PP6's pivotal role in melanoma involves regulating key pathways linked to cell proliferation, survival, and invasion, making it a potential target for intervention. Leveraging our robust preliminary data, in the current proposal we aim to comprehensively grasp the assembly of PP6 holoenzyme and its recruitment of substrates through rigorous structural and cellular studies.

B. Positions and Honors

Positions and Employment

2021-Current	Assistant Professor (In Residence), Laboratory of Drs. Wolfgang Peti & Rebecca Page, University of Connecticut School of Medicine, Farmington, CT
2019 -2020	Scientist III. University of Arizona, Tucson, AZ
2013 - 2019	Postdoctoral Research Associate, Laboratory of Dr. Andrew S Kraft, Hollings Cancer Center, Charleston, SC and University of Arizona Cancer Center, Tucson, AZ
2010 - 2012	Teaching Assistant, Pharmaceutical Sciences, North Dakota State University, Fargo, ND
2008 - 2013	Graduate Assistant, Pharmaceutical Sciences, North Dakota State University Fargo, ND
2006 - 2007	Teaching Assistant, Chemistry and Biochemistry, University of Detroit Mercy, Detroit, MI.
2005 - 2008	Graduate Assistant, Chemistry and Biochemistry, University of Detroit Mercy, Detroit, MI
2004 - 2005	Senior Scientific Content Developer. (Medical Writer), Foot Prints, Hyderabad, India
2003	Trainee: Industrial in-plant training at Cadila Pharmaceuticals, Ahmedabad, India

Honors

2023	Invited talk on "Cryo-EM structures of PP2A:B55-inhibitor complexes: substrate recruitment mechanisms". EMBO workshop – Signal regulation by protein phosphatases: mechanisms and pathways
2022-Current	Early Career Reviewer at Journal of Biological Chemistry
2022-Current	UConn Health Biomedical Science PhD Program Admissions Committee member
2021	Invited talk/Panel Discussion on "Making PI3K Inhibitors Work in the Clinic" at Inaugural virtual PI3K Pathways Summit
2020	PIM kinase inhibitors block the growth of primary T-cell acute lymphoblastic leukemia: Resistance pathways identified by network modeling analysis – Featured in Highlights of Molecular Cancer Therapeutics journal -September 2020 | Volume 19 | Issue 9
2018	Invited/selected for a short talk in a plenary session at the AACR Special Conference on Targeting PI3K/mTOR Signaling, Boston, MA.
2018	Scholar-in-Training Award supported by Aflac, Inc. AACR Special Conference on "Targeting PI3K/mTOR Signaling", Boston, MA.
2016	ASH Abstract achievement award, 58th ASH Annual Meeting & Exposition, San Diego, CA.
2014	Post-doctoral scholarship from Abney and Associates foundation, Anderson, SC
2013	Graduate Student Travel Award from ASPET (American Society for Pharmacology & Experimental Therapeutics)
2013	Young Investigator Award from SEBM (Society for Experimental Biology and Medicine)
2012-2013	Darryle and Clare Schoepp Graduate Research Scholarship, by the NDSU College Scholarship Recognition Committee

2012-2013	Treasurer of AAPS-NDSU Student Chapter
2011-2013	ND EPSCoR-DDA Award at NDSU (North Dakota Experimental Program to Stimulate Competitive Research - Doctoral Dissertation Assistantship)
2010	Certificate from “The Honor Society of Phi Kappa Phi”, North Dakota State University

Other Experience and Professional Memberships

2016-Present	Manuscript Reviewer for Molecular Cancer Therapeutics, Journal of Biological Chemistry, Oncotarget, Blood and Lymphatic Cancer, Leukemia Research, OncoTargets and Therapy, International Journal of Hematology, Blood Reviews, Cancers, Life, International Journal of Molecular Sciences and Hematology Reports
2016-Present	Editorial member of Austin Journal of Medical Oncology and Cancer Clinical Research Reports
2016-2019	American Society of Hematology (ASH)
2011-Present	American Association for Cancer Research (AACR)

C. Contribution to Science

(i) **High resolution Cryo-EM structures of two PP2A:B55-inhibitor complexes:** To gain knowledge and further understand the molecular insights into the substrate specificity of Serine/Threonine Phosphatases, I joined the laboratory of Drs. Wolfgang Peti and Rebecca Page as an Assistant Professor (In Residence) at UConn Health, Molecular Biology and Biophysics Department. PP2A is one of the most abundant serine/threonine phosphatases in eukaryotic cells. PP2A activity is deregulated in various cancers, leading to abnormal cell growth, a hallmark of malignancy. PP2A:B55 function is regulated by two established intrinsically disordered protein (IDP) inhibitors, FAM122A and ARPP19. Despite advances in defining how regulators and substrates are recruited by other phosphatases, the substrate recruitment by PP2A:B55 holoenzyme is poorly understood, limiting our knowledge on PP2A:B55 mediated cell signaling. By leveraging cryo-EM, mutagenesis coupled with fluorescence polarization binding, inhibition and pulldown assays, we identified the key interaction motif(s) that promote PP2A:B55-Inhibitor(s) complexes formation. Furthermore, through NMR and cell-based competition assays we show that PP2A:B55 uses multiple interaction surfaces to recruit its regulators/substrates, allowing for high specificity. By combining our structural data with cBioPortal analysis, we also demonstrated how cancer mutations within these inhibitors lead to PP2A:B55 dysregulation in various cancers.

Padi SKR, Vos MR, Godek RJ, Fuller JR, Kruse T, Hein JB, Nilsson J, Kelker MS, Page R and Peti W. **(2023)** Cryo-EM structures of PP2A:B55-FAM122A and PP2A:B55-ARPP19. **Nature**. https://doi.org/10.1038/s41586-023-06870-3. (Published 20 December 2023).

PadiSKR, Page R, Peti W. Cryo-EM structure of PP2A:B55-inhibitor complexes highlight substrate recruitment mechanisms. Oral presentation at **EMBO workshop** – Signal regulation by protein phosphatases: mechanisms and pathways**.** August 6-10, 2023; Copenhagen, Denmark.

(ii) **Targeting PIM kinases to overcome drug resistance mechanisms:** Researchers have provided a rationale and basis for co-targeting PIM kinases with inhibitors of PI3K/mTOR/AKT, JAK/STAT, MYC, stemness, and RNA Polymerase I transcription, along with other treatments, including androgen deprivation therapy, radiotherapy, chemotherapy, and immunotherapy. Such combined approaches could potentially be used as neoadjuvant therapies, limiting the development of resistance to treatments or sensitizing cells to other therapeutics. We have identified a phosphorylation-dependent mechanism that controls mTORC1 activity in which Pim and AKT kinases, 2 enzymes with increased activity in cancer phosphorylate DEPDC5, a member of the GATOR1 complex that senses cellular amino acid levels. The critical nature of this substrate to the activity of these protein kinases is demonstrated by the fact that deletion or mutation of DEPDC5 partially blocks the ability of Pim and Pim plus AKT inhibitors to suppress tumor cell growth. Thus, protein kinases regulate the amino acid sensing cascade to control mTORC1 activity and tumor cell growth.

Padi SKR, Singh N, Bearss JJ, Olive V, Song JH, Cardó-Vila M, Kraft AS, and Okumura K. (**2019**) Phosphorylation of DEPDC5, a component of the GATOR1complex, releases inhibition of mTORC1 and promotes tumor growth. **Proceedings of the National Academy of Sciences.** PMID: 31548394 *(cited by 18 articles)*

Song JH, Singh N, Luevano LA, **Padi SKR**, Okumura K, Olive V, Black SM, Warfel NA, Goodrich DW, and Kraft AS. (**2018**) Mechanisms behind resistance to PI3K Inhibitor treatment induced by the PIM kinase. **Molecular Cancer Therapeutics.** PMID: 30190422 *(cited by 43 articles)*

Bearss JJ#, **Padi SKR**#, Singh N, Cardó Vila M, Song JH, Mouneimne G, Fernandes N , Li Y, Harter MR, Gard JMC, Cress A, Peti W, Nelson A, Buchan JR, Kraft AS and Okumura K. (**2021**) EDC3 phosphorylation regulates growth and invasion through controlling P-body formation and dynamics. **EMBO Reports**. PMID: 33586867 (# Co-first author, *cited by 15 articles*)

Gnawali GR, Okumura K, Perez K, Gallagher R, Wulfkuhle J, Petricoin EF, **Padi SKR**, Bearss J, He Z, Wang W, Kraft AS. (**2022**) Synthesis of 2-oxoquinoline derivatives as dual PIM and mTORC protein kinase inhibitors. **Medicinal Chemistry Research**. 31, pages1154–1175.

(iii) **PIM kinases: A potential therapeutic target in T-cell Acute Lymphoblastic Leukemia:** For my Post-doctoral training, I joined the laboratory of Dr. Andrew S Kraft, Professor and Director Emeritus of the University of Arizona Cancer Center, where I got an opportunity to investigate the role of PIM oncogenic kinases in hematopoietic and solid tumor malignancies. Dr. Kraft's lab has been a leader in the study of PIM kinases, and successfully synthesized and developed a PIM kinase inhibitor (SMI-4a), which validated PIM as a relevant therapeutic target in cancer. Under the mentorship of Dr. Kraft, I have made a novel observation that PIM1 is overexpressed in early T cell precursor (ETP)-ALL, a subset of T-ALL, which are shown to be chemo-refractory due to their heterogeneous nature along with activating mutations. This, together with our observation that combining PIM inhibitors with Ponatinib, an oral multi-targeted tyrosine kinase inhibitor induces substantial apoptosis of ETP-ALL cells, will enhance the development of a new therapeutic option towards significantly blocking the T-ALL growth in pre-clinical setting, and in the future for children and adults with difficult to treat T-ALL.

Padi SKR, Luevano LA, An N, Pandey R, Singh N, Song JH, Aster JC, Yu XZ, Mehrotra S, and Kraft AS. (**2017**) Targeting the PIM Protein Kinases for the Treatment of a T-cell Acute Lymphoblastic Leukemia Subset. **Oncotarget**. doi: 10.18632/oncotarget.16320. PMCID: PMC5444737 (*cited by 42 articles*)

Song JH, **Padi SKR**, Luevano LA, Minden MD, DeAngelo DJ, Hardiman GT, Ball LE, Warfel NA, and Kraft AS. (**2016**) Insulin Receptor Substrate 1 Is a Substrate of the Pim Protein Kinases. **Oncotarget**. doi: 10.18632/oncotarget.7918. PMCID: PMC49914444 (*cited by 29 articles*)

Singh N, **Padi SKR**, Bearss JJ, Pandey R, Okumura K, Beltran H, Song JH, Kraft AS, and Olive V. **(2020)** PIM protein kinases regulate the level of the long noncoding RNA H19 to control stem cell gene transcription and modulate tumor growth. **Molecular Oncology.** PMID: 32146726 *(cited by 22 articles)*

Lim JT, Singh N, Leuvano LA, Calvert VS, Petricoin EV, Teachey D, Lock R, Padi M, Kraft AS*, and **Padi SKR*. (2020)** PIM kinase inhibitors block the growth of primary T-cell acute lymphoblastic leukemia: Resistance pathways identified by network modeling analysis. ***Corresponding author**; **Molecular Cancer Therapeutics**. PMID: 3275338 *(cited by 6 article)*

(iv) **Epigenetic mechanisms and Drug Resistance:** As a PhD student under the guidance of Dr. Bin Guo, my research was focused on the understanding of how epigenetic mechanisms contribute to drug resistance mainly in colon cancer and prostate cancer. We identified the novel targets (miR-627 & JMJD1A) downstream of vitamin D, which can be used to design a new therapy for colon cancer and these results were published in "Gastroenterology" (**impact factor 29.4**), the most cited journal in the field of gastrointestinal diseases. This work resulted in an NIH R01 grant to Dr. Bin Guo "RNA Therapeutics for Targeted Treatment of Colon Cancer" (R01 CA186100-01A1; 5/19/2015–4/30/2020; $1,351,550).

Bhatnagar N*, Li X*, **Padi SKR***, Zhang Q, Tang MS, Guo B. (**2010**) Downregulation of miR-205 and miR-31 confers resistance to chemotherapy-induced apoptosis in prostate cancer cells. **Cell Death and Disease**, 1, e105; doi:10.1038/cddis.2010.85. PMCID: PMC3004480 (* Co-first author*, cited by 232 articles*)

Padi SKR, Guo B. MiR-627 mediates the epigenetic mechanism of vitamin D in suppression of colon cancer growth both in vitro and in vivo. **EB conference**, **2013**. The **FASEB** Journal 27 (1_supplement), 1104.5.

Padi SKR, Guo B. MiR-627 and histone demethylase JMJD1A as new therapeutic targets in colon cancer. **AACR**, **2012**. **Cancer Research** 2012;72(8 Suppl): Abstract nr 4739.

Padi SKR, Zhang Q, Rustum YM, Morrison C, Guo B. (**2013**) MicroRNA-627 Mediates the Epigenetic Mechanisms of Vitamin D to Suppress Proliferation of Human Colorectal Cancer Cells and Growth of Xenograft Tumors in Mice. **Gastroenterology**. doi: 10.1053/j.gastro.2013.04.012. PMCID: PMC3722307 (Frist author, *cited by 130 articles*)

(v) **Collaboration work with other researchers at North Dakota State University.**

Zhang Q, **Padi SKR**, Tindall DJ, Guo B. (**2014**) Polycomb protein EZH2 suppresses apoptosis by silencing the proapoptotic miR-31. **Cell Death and Disease**. doi:10.1038/cddis.2014.454. PMCID: PMC4237267 (*cited by 89 articles*)

Kulkarni PS, Haldar MK, Nahire RR, Katti P, Ambre AH, Muhonen WW, Shabb JB, **Padi SKR**, Singh RK, Borowicz PP, Shrivastava DK, Katti KS, Reindl K, Guo B, Mallik S. (**2014**). MMP-9 Responsive PEG Cleavable Nanovesicles for Efficient Delivery of Chemotherapeutics to Pancreatic Cancer. **Molecular Pharmaceutics**. doi: 10.1021/mp500108p. PMCID: PMC4096225 (*cited by 105 articles*)

Singh RK, Cho K, **Padi SKR**, Yu J, Haldar M, Mandal T, Yan C, Cook G, Guo B, Mallik S, Srivastava DK. (**2015**) Mechanism of N-Acylthiourea-mediated Activation of Human Histone Deacetylase 8 (HDAC8) at Molecular and Cellular Levels. **The Journal of Biological Chemistry**. doi: 10.1074/jbc.M114.600627. PMCID: PMC4358293 (*cited by 23 articles*)

Complete List of Published Work:

https://www.ncbi.nlm.nih.gov/myncbi/1PYwZlZGVjYc9k/bibliography/public/