BIOGRAPHICAL SKETCH

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NAME: Tesmer, John Joseph Grubb

eRA COMMONS USER NAME: johntesmer

POSITION TITLE: Walther Distinguished Professor of Cancer Structural Biology

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Rice University, Houston TX	BA	06/1990	Biochemistry & English
Purdue University, West Lafayette IN	PHD	12/1995	Biological Sciences
University of Texas Southwest Medical Center at Dallas, Dallas TX	postdoc	09/1999	Structural Biology/ Signal Transduction

A. Personal Statement

My lab is best known for biophysical studies of G protein-coupled receptor (GPCR) signaling pathways. To this end, we use X-ray crystallography, single particle cryo-electron microscopy (cryo-EM), hydrogen-deuterium exchange mass spectrometry, and small-angle X-ray scattering to tease apart the mechanisms by which the proteins in GPCR cascades interact and transfer information. A key focus has been the structure and function of GPCR kinases (GRKs), a family of seven enzymes that selectively phosphorylate activated GPCRs, triggering their uncoupling from G proteins and promoting receptor internalization. Our lab also leverages biophysical information to accelerate the design of compounds targeting the GRKs, whose overexpression in diseases such as heart failure, cardiac hypertrophy, and cancer is linked to poor patient outcome. Seminal achievements in our lab concerning the GRKs were the first structure of a GRK (*Science*, 2003), our recent landmark cryo-EM structure of rhodopsin kinase in complex with rhodopsin (*Nature*, 2021), and the discovery of selective and potentially therapeutic inhibition of GRK2 by the FDA-approved drug paroxetine and its derivatives (*ACS Chem Biol* 2012; *Sci Trans Med*, 2015). Our GRK work recently segued into studies of atypical chemokine receptor 3 (ACKR3), an important cancer target that exhibits intrinsic signaling bias towards arrestins and GRKs and that scavenges the chemokine ligand for CXCR4, its close homolog and physiological partner. To this end, we have characterized ligand activated ACKR3 by cryo-EM and revealed structural features likely responsible for its intrinsic bias (*Sci Adv*, 2022), and most recently have described the molecular basis for how different isoforms of arrestin and GRKs interact with ACKR3 (*Nature*, 2025). We also investigate the structure and function of heterotrimeric G protein effector enzymes, in particular GPCR-regulated RhoGEFs, essential for neutrophil chemotaxis as well as tumor growth and cancer metastasis. Key papers thus far describe the $G\alpha_q$–p63RhoGEF–RhoA complex (*Science*, 2007) and cryo-EM structures of the P-Rex1–G$\beta\gamma$ complex (*Sci Adv*, 2019; *eLife*, 2004), the PI3Kγ–G$\beta\gamma$ complex (*NSMB*, 2024), and the adenylyl cyclase 5–G$\beta\gamma$ complex (*NSMB*, 2024). I have contributed to over 158 publications (h-index of 49 in Scopus as of Apr 2025). I have enjoyed nearly continuous extramural support from the NIH since 2003 and been recognized with several prestigious awards (ASPET J. Jacob Abel Award, ASBMB Young Investigator Award, AAAS Fellow, Distinguished Professor, ASPET Fellow, and ASPET Ruffolo Career Achievement Award) consistent with a high level of productivity, impact, and contributions to the scientific community.

I have a strong commitment to scientific mentoring and encouraging the participation of talented junior scientists from any background. I have worked with 18 post-doctoral fellows, mentored 33 PhD/MS students, and trained over 75 undergraduate researchers. The current members of my lab hail from 8 different nations and we have established a supportive environment for those at all language and technical sufficiency levels. All trainees take the required courses in lab safety, RCR, and statistics, and I reinforce these principles and ethical practices routinely during our weekly group and 1-on-1 meetings. For example, I recognize the importance of depositing models, primary diffraction data, and EM maps in public structural databases, and we readily share our reagents with others upon reasonable request. I ensure that my lab adheres to current standards in statistical reporting and significance testing, and uses rigorous practices such as assaying multiple preparations of proteins to

support key results. I work with all graduate students to formulate an IDP for their professional development on an annual basis. Proficiency in independent grant writing is also a high priority for us. To this end, five of my graduate students have received American Heart Association (AHA) pre-doctoral fellowships, and one an F31 fellowship. Seven of my post-doctoral trainees received AHA post-doctoral fellowships, one an American Cancer Society fellowship, and one an F32 fellowship. Most of my alumni have remained in science, with 10 now employed as academic faculty and 4 as senior scientists in the private sector. I have served on multiple AHA and NIH study sections and ASPET's governing council, as an editor for *Journal of Biological Chemistry*, and as a Deputy Editor and now Editor-in-Chief for the ASPET journal *Molecular Pharmacology*. I am especially dedicated to biophysics education and outreach. Currently, I am the Director of the Purdue University NIGMS T32 Molecular Biophysics Training Grant, which provides advanced coursework and professional development to 7 outstanding biophysics graduate students each year.

Ongoing projects that I would like to highlight include:

R01 CA254402
Tesmer/Handel (PI)
6/1/20 - 5/31/26 (NCE)
Regulation of the metastasis promoting chemokine receptor ACKR3 by GPCR kinases, Gβγ and arrestins

R01 HL071818
Tesmer (PI)
4/1/20 - 3/31/25 (NCE)
Structure, function, and inhibition of G protein-coupled receptor kinases

R01 CA294770
Myers/Tesmer/Atwood (Co-PIs)
6/1/25 – 5/31/29 (4th percentile; funding delayed)
Elucidating the functional role and therapeutic potential of GPCR kinase 2 in primary and therapy-resistant basal cell carcinoma

T32 GM132024
Tesmer (PI)
7/1/25 – 6/31/30 (priority score 29; funding decision delayed)
Purdue University Molecular Biophysics Training Program

B. Positions, Scientific Appointments, and Honors

Positions

2021 -	Distinguished Professor, Purdue University
2019 -	Leader: Targets, Structures and Drugs Program (Purdue Institute for Cancer Research)
2018 -	Professor, Department of Medicinal Chemistry and Molecular Pharmacology (courtesy appt)
2017 -	Walther Professor in Cancer Structural Biology, Biological Sciences, Purdue University
2017 - 2019	Adjunct Professor, Departments of Pharmacology and Biological Chemistry, U of Michigan
2011 - 2017	Professor, Department of Pharmacology, University of Michigan
2011 - 2017	Professor, Life Sciences Institute, University of Michigan
2005 - 2011	Research Associate Professor, Life Sciences Institute, University of Michigan
2005 - 2011	Associate Professor, Department of Pharmacology, University of Michigan
1999 - 2005	Assistant Professor, Department of Chemistry, University of Texas at Austin
1996 - 1999	Howard Hughes Postdoctoral Fellow, U of Texas Southwestern Medical Center at Dallas

Scientific Appointments

2023-2027	Member, NIH TWD-B Study Section
2023-2025	Editor-in-Chief, *Molecular Pharmacology*
2017-2019	Treasurer Elect, Treasurer, & Past Treasurer-ASPET Council
2016-2020	Member, NIH MIST Study Section
2012-2014	Chair Elect, Chair, & Past Chair ASPET Molecular Pharmacology Division
2012-	Editorial Board of *Molecular Pharmacology;* Associate Editor (2014-22); Deputy Editor (2017-22)
2012	Chair, Gordon Research Conference on Phosphorylation & G-Protein Mediated Signaling Networks, University of New England
2012	NIEHS Division of Intramural Research Board of Scientific Counselors Review
2010-2016	ASPET Awards Committee

2009-2019 ASPET Molecular Pharmacology Division Executive Committee
2007-2017 Life Sciences-CAT (Advanced Photon Source) management board (UM representative)
2007-2012 Editorial Board of *Journal of Biological Chemistry*
2006- American Heart Association grant review: BCMB 3 Study Section (2006), PC1 and PC3 (2013-4), Co-Chair PC1 (2015-6), Chair PC1 (2017), Chair Fellowship BC (2018)
2001-2005 Institutional Biosafety Committee, UT Austin, TX

Honors

2025 Robert R Ruffolo Career Achievement Award (ASPET)
2025 Keynote Lecture, FASEB SRC GRK/Arrestin Meeting
2024 Purdue Institute for Cancer Research (PICR) Career Research Achievement Award
2024 Named Fellow of American Society for Pharmacology and Experimental Therapeutics (FASPET)
2022 Keynote Lecture, Phosphorylation and G-Protein Mediated Signaling Networks GRC
2013 Named Fellow of the American Association for the Advancement of Science (FAAAS)
2012 - 2017 Cyrus Levinthal Collegiate Professorship in the Life Sciences (U. of Michigan)
2010 American Society for Biochemistry and Molecular Biology Young Investigator Award
2009 John J. Abel Award (American Society for Pharmacology and Experimental Therapeutics)
2008 Basic Science Research Award (U. Michigan Medical School)
2004 U. of Texas at Austin College of Natural Sciences Teaching Excellence Award
2002 Cottrell Scholar Award (Research Corporation)
2000 American Heart Association Texas Affiliate Lyndon Baines Johnson Research Award
1995 H. E. Umbarger Outstanding Graduate Student Award
1994 - 1995 Purdue Research Foundation Grant
1990 - 1993 N.S.F. Predoctoral Fellowship

C. Contributions to Science

a. Molecular Basis of Heterotrimeric G Protein Function. Signal transduction conveyed from G protein-coupled receptors (GPCRs) via heterotrimeric G proteins is one of the classic paradigms of hormone action, wherein extracellular signals lead not only to transient changes in the concentrations of intracellular second messengers but also to sustained changes such as chemotaxis, cell growth, and metastasis. As a post-doctoral fellow with Stephen Sprang, I determined the atomic structure of the RGS4-Gαi1 complex (Tesmer *et al. Cell* 1997), the first of a regulator of G protein signaling (RGS) protein and of an RGS protein in complex with a Gα subunit. I followed this with crystal structures of Gαs alone (Sunahara *et al. Science* 1997) and bound to the catalytic domains of adenylyl cyclase (Tesmer *et al. Science* 1997; Tesmer et al. *Science* 1999), the first of a Gα-effector enzyme complex. As an independent investigator, my lab has focused on effector enzymes responsive to Gβγ and Gαq (3,4), which are key regulators of cardiovascular function and cell growth/transformation. Current projects in the lab include investigating how Gβγ subunits regulate full-length mammalian adenylyl cyclase 5 (3) and phosphoinositide 3-kinase γ (4).

1. Lutz S, Shankaranarayanan A, Coco C, Ridilla M, Nance MR, Vettel C, Baltus D, Evelyn CR, Neubig RR, Wieland T, **Tesmer JJ**. Structure of Gαq-p63RhoGEF-RhoA complex reveals a pathway for the activation of RhoA by GPCRs. *Science* 2007, **318**:1923-7. PMID: 18096806.
2. Lyon AM, Dutta S, Boguth CA, Skiniotis G, **Tesmer JJ**. Full-length Gαq-phospholipase C-β3 structure reveals interfaces of the C-terminal coiled-coil domain. *Nat Struct Mol Biol.* 2013, **20**:355-62. PMCID: PMC3594540.
3. Yen Y-C, Li Yong, Chen C-L, Klose T, Watts VJ, Dessauer CW, and Tesmer JJG. Isoform specific regulation of adenylyl cyclase 5 by Gβγ. *Nat Struct Mol Biol.* 2024, **31**:1189-1197. doi: 10.1038/s41594-024-01263-0. PMCID: PMC11329361
4. Chen C-L, Ravala SK, Yen Y-C, and **Tesmer JJG**. Structural and mechanistic basis for Gβγ-mediated activation of phosphoinositide 3-kinase γ. *Nat Struct Mol Biol*. 2024, **31**:1198-1207. PMCID: PMC11329362.

b. Structure and Function of G Protein-Coupled Receptor Kinases (GRKs). The ~800 GPCRs in the human genome are regulated by a family of seven kinases that inhibit signaling by activated GPCRs, ensuring a return to homeostasis. In 2003, my lab published the structure of the GRK2-Gβγ complex, the first of a GRK and the first of Gβγ subunits in complex with an effector (Lodowski *et al. Science* 2003). In 2005, the lab reported the structure of the $G\alpha_q$-GRK2-Gβγ complex (1), providing a snapshot of activated Gα and Gβγ

subunits at the membrane as they engage a common target. The work also yielded the first atomic structure of $G\alpha_q$. The lab went on to characterize GRKs from other subfamilies: GRK6, GRK1 (rhodopsin kinase), and GRK5. These structures helped to identify sites that form the docking site for activated GPCRs and anionic phospholipids. Most recently, we determined the cryo-EM structure of the rhodopsin–GRK1 (rhodopsin kinase) complex, the first of a GPCR engaged by a GRK (2). Current efforts are directed towards determining structures of rhodopsin and ACKR3 with other GRKs to refine our models of how GRK selectivity for receptors is achieved. To this end, we have determined the structures of a series of agonist bound complexes of ACKR3 (3) and have defined the molecular consequences of phosphorylation barcoding on arrestin isoform configuration by different GRK subfamilies using cryo-EM (4).

1. Tesmer VM, Kawano T, Shankaranarayanan A, Kozasa T, **Tesmer JJG**: Snapshot of activated G proteins at the membrane: the $G\alpha_q$-GRK2-$G\beta\gamma$ complex. *Science* 2005, **310**: 1686-1690. PMID: 16339447.
2. Chen Q, Plasencia M, Li Z, Mukherjee S, Patra D, Chen C-L, Klose T., Yao X-Q, Kossiakoff AA, Chang L, Andrews PC, **Tesmer JJG**: Structures of rhodopsin in complex with G-protein-coupled receptor kinase 1. *Nature* 2021, **595**:600-605. PMC8607881.
3. Yen YC, Schafer CT, Gustavsson M, Eberle SA, Dominik PK, Deneka D, Zhang P, Schall TJ, Kossiakoff AA, **Tesmer JJG**, Handel TM. Structures of atypical chemokine receptor 3 reveal the basis for its promiscuity and signaling bias. *Sci Adv*. 2022, **8**:eabn8063. PMC9278869.
4. Chen Q, Schafer CT, Mukherjee S, Kossiakoff AA, Wang K, Gustavsson M, Fuller JR, Tepper K, Lamme TD, Aydin Y, P Agarwal, Terashi G, Yao X-Q, Kihara D, Kossiakoff AA, Handel TM, and **Tesmer JJG**. Effect of phosphorylation barcodes on arrestin binding to a chemokine receptor *Nature*, 2025, https://doi.org/10.1038/s41586-025-09024-9.

c. GPCR-Linked Rho Guanine Nucleotide Exchange Factors (RhoGEFs). Sustained changes in cell behavior induced by GPCRs typically involve modulating the actin cytoskeleton and gene transcription via RhoGEFs. These enzymes play a central role in chemotaxis, tumor growth and metastasis by activating various members of the Rho GTPase family. In 2004, my lab reported atomic structures of the catalytic domains of leukemia-associated RhoGEF (LARG) alone and in complex with RhoA (Kristelly *et al. JBC* 2004), and in 2006, the structures of the oncogenic $G\alpha_{12}$ and $G\alpha_{13}$ subunits that regulate the activity of LARG and closely related RhoGEFs (Kreutz *et al. Biochemistry* 2006). In 2007, the lab resolved the structure of the $G\alpha_q$-p63RhoGEF-RhoA ternary complex, capturing a snapshot of a novel $G\alpha_q$ signaling pathway implicated in the development of cardiac hypertrophy (Lutz *et al. Science* 2007). More recently, we have been characterizing the Rac1 specific enzyme, P-Rex1 (1-3), a $G\beta\gamma$-regulated RhoGEF that plays a key role in neutrophil function and in metastasis of breast and prostate cancer, and Trio (4), a close relative of p63RhoGEF responsible for tumor growth in uveal melanoma.

1. Cash JN, Davis EM, **Tesmer JJG**: Structural and biochemical characterization of the catalytic core of the metastatic factor P-Rex1 and its regulation by PtdIns(3,4,5)P_3. *Structure* 2016, **24**:730-40. PMC4860252.
2. Cash JN, Urata S, Li S, Ravala S, Avramova L, Gutkind JS, **Tesmer JJG**, Cianfrocco MA: Cryo-electron microscopy structure and analyses of the P-Rex1–$G\beta\gamma$ complex signaling scaffold. *Sci Adv.* 2019, **5**: eaax8855. PMC6795519.
3. Ravala SK, Adame S, Li S, Chen C-L, Cianfrocco MA, Gutkind JS, Cash JN, and **Tesmer JJG**. Structural and dynamic changes in P-Rex1 upon activation by PIP_3 and inhibition by IP_4. *Elife* 2024, **12**:RP92822. doi: 10.7554/eLife.92822. PMC11290822.
4. Bandekar SJ, Arang N, Tang BA, Tully ES, Barton BL, Li S, Gutkind JS, and **Tesmer JJG**: Structure of the C-terminal guanine exchange factor module of Trio in an autoinhibited conformation reveals its oncogenic potential. *Sci Signal*. 2019, **12**: pii: eaav2449. PMC6519057

d. Molecular Basis of Phospholipase Activation. Phospholipases are potent esterases whose activity must be tightly regulated to prevent lipid hydrolysis at the wrong membranes and/or in the absence of appropriate extracellular signals. The phospholipase Cβ family responds to extracellular signals that act on GPCRs by hydrolyzing PIP_2 to produce the second messengers diacylglycerol and IP_3. My lab defined new $G\alpha_q$-responsive autoinhibitory elements in the C-terminal domain of PLCβ and determined the first full-length structure of PLCβ in complex with $G\alpha_q$ (see section **a** above). The LPLA2/LCAT family of phospholipases transfers the *sn*2 acyl chain from lipids such as lecithin to acceptor lipids such as ceramide or cholesterol.

These closely related enzymes are important for lung surfactant catabolism and reverse cholesterol transport to the liver via HDL, respectively, and are both targets for biotherapeutic development. My lab determined crystal and negative stain EM structures for both LPLA2 and LCAT (1,2,3,4) and has studied the mode of action of small molecular activators (3) that could be used to treat acute coronary syndrome and/or fatal genetic diseases that inhibit LCAT activity, such as familial LCAT deficiency.

1. Glukhova A, Hinkovska-Galcheva V, Kelly R, Abe A, Shayman JA, **Tesmer JJ**. Structure and function of lysosomal phospholipase A2 and lecithin:cholesterol acyltransferase. *Nat Commun.* 2015, **6**:6250. PMC4397983.
2. Hoadley-Manthei K, Ahn J, Glukhova A, Yuan W, Larkin C, Manett TD, Chang L, Shayman JA, Axley MJ, Schwendeman A, Shayman JA, **Tesmer JJG**: A retractable lid in lecithin:cholesterol acyltransferase provides a structural mechanism for activation by apolipoprotein A-I. *J. Biol. Chem.* 2017, **292**:20313-20327. PMC5724016
3. Manthei KA, Yang S-M, Baljinnyam B, Glukhova A, Chang L, Yuan W, Freeman LA, Shen M, Maloney DJ, Schwendeman A, Remaley AT, Jadhav A, and **Tesmer JJG**: Molecular basis for activation of lecithin:cholesterol acyltransferase (LCAT) by a high affinity chemical probe. *Elife*, 2018, **7**: pii: e41604. PMC6277198
4. Manthei KA, Patra D, Wilson CJ, Fawaz MV, Piersimoni L, Shenkar J, Yuan W, Andrews PC, Engen JR, Schwendeman A, Ohi MD, **Tesmer JJG**. Structure of lecithin:cholesterol acyltransferase bound to high density lipoprotein particles. *Commun. Biol.* 2020, **3**:28. PMC6962161

e. Identification and Rational Design of Small Molecule Probes. We also use biophysics to accelerate the discovery of selective small molecule agents that can be used to probe the above signaling cascades in more physiological contexts, or that can serve as leads for drug development. Our most advanced work in this realm involves GRK inhibitors. GRK2 subfamily inhibitors have potential applications ranging from treatment of congestive heart failure to inhibition of arterial and renal plaque formation. GRK5 subfamily inhibitors are expected to have utility in treatment of cardiac hypertrophy and cancer. In 2012, we identified the selective serotonin reuptake inhibitor paroxetine (Paxil) as a selective GRK2/3 inhibitor, which was later shown to reverse heart malfunction in mice subjected to infarction (1). We have identified additional potent chemical scaffolds with distinct selectivity profiles for the various GRKs and are developing these as leads with our medicinal chemistry collaborators (2,3,4).

1. Schumacher SM, Gao E, Zhu W, Chen X, Chuprun JK, Feldman AM, **Tesmer JJ**, Koch WJ. Paroxetine-mediated GRK2 inhibition reverses cardiac dysfunction and remodeling after myocardial infarction. *Sci Transl Med*. 2015, **7**:277ra31. PMC4768806
2. Waldschmidt HV, Homan KT, Cato MC, Cruz-Rodríguez O, Cannavo A, Wilson MW, Song J, Cheung JY, Koch WJ, **Tesmer JJG**, Larsen SD: Structure-based design of highly selective and potent G protein-coupled receptor kinase 2 inhibitors based on paroxetine. *J. Med. Chem*. 2017, **60**:3052-3069. PMC5641445
3. Rowlands RA, Chen Q, Bouley RA, Avramova LV, **Tesmer JJG**, White AD: Generation of highly selective, potent, and covalent G protein-coupled receptor kinase 5 inhibitors. *J. Med. Chem.* 2021, **64**:566-585. PMCID: PMC7909074
4. Chen Y, Sonawane A, Manda R, Gadi RK, **Tesmer JJG**, Ghosh AK. Development of a new class of potent and highly selective G protein-coupled receptor kinase 5 inhibitors and structural insight from crystal structures of inhibitor complexes. *Eur. J. Med. Chem*. 2024 **264**: 115931. PMCID: PMC10841647

Complete List of Published Work:

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