BIOGRAPHICAL SKETCH

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NAME: Lee, Seok-Yong

eRA COMMONS USER NAME (credential, e.g., agency login): SYLEE1

POSITION TITLE: Professor

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

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
Yonsei University, Seoul, Korea	B.S.	05/1998	Biochemistry
University of California at Berkeley, Berkeley, California	Ph.D.	05/2003	Biophysics
Rockefeller University, New York City, New York	Postdoctoral fellow	07/2009	Structural Biology and Biophysics

A. Personal Statement

I have over 20 years of experience in the field of membrane protein structural biology and biophysics. During my independent career at Duke University School of Medicine, my laboratory has carried out structural and mechanistic studies of membrane transport proteins that are important in many different physiological processes using cryo-electronmicroscopy (cryo-EM), X-ray crystallography, electrophysiology, and various biophysical methods. My lab has contributed significantly to our molecular-level understanding of 1) the sensations of heat, capsaicin (spiciness), menthol (coolness), and wasabi (pungent compounds) in humans, 2) drug and metabolite uptake in humans, and 3) lipid transport critical for the bacterial cell wall synthesis. These studies have not only significantly advanced our fundamental understanding of these mechanisms, but also contribute information for the future development of analgesic, antiviral, anticancer and antibiotic agents. In summary, I have demonstrated expertise and productivity in the area of structural and mechanistic studies of integral membrane proteins and am well prepared to carry out the proposed research program.

Citations:

a) Ying Yin, Feng Zhang, Shasha Feng, Kevin John Butay, Mario J. Borgnia, Wonpil Im, **Seok-Yong Lee***, Activation mechanism of the mouse cold-sensing TRPM8 channel by cooling agonists and PIP_2. *Science*, 2022. 378, add1268. PMCID: PMC9795508

b) Nicholas Wright, Justin Fedor, Han Zhang, Pyeonghwa Jeong, Yang Suo, Jiho Yoo, Jiyong Hong, Wonpil Im, **Seok-Yong Lee***. "Methotrextate recognition by the human reduced folate carrier SLC19A1". *Nature,* 2022. Sep;609(7929):1056-1062. PMCID: PMC9822521

c) Yang Suo, Nicholas Wright, Hugo Guterres, Justin Fedor, Kevin John Butay, Mario J. Borgnia, Wonpil Im, **Seok-Yong Lee***. Molecular basis of polyspecific drug and xenobiotic recognition by OCT1 and OCT2. *Nat. Struct. Mol. Biol.*, 2023 Jul;30(7):1001-1011. PMID:37291422

d) Nicholas Wright, Feng Zhang, Yang Suo, Linyang Kong, Ying Yin, Kedar Sharma, Mario Borgnia, Wonpil Im, **Seok-Yong Lee***. Antiviral drug recognition and elevator-type transporter motions of CNT3. *Nat. Chem. Biol.* 2024 In press, doi:10.1038/s41589-024-01559-8

B. Positions, Scientific Appointments, and Honors

Positions and Scientific Appointments

2023.7 – Present George Barth Geller Distinguished Professor of Molecular Biology, Duke University

2020.7 – Present Professor of Biochemistry, Duke University School of Medicine, Durham, NC
2016.7 – 2020.6 Associate Professor of Biochemistry, Duke University School of Medicine, Durham, NC.
2009.9 – 2016.6 Assistant Professor of Biochemistry, Duke University School of Medicine, Durham, NC.
2003.7 – 2009.7 Postdoctoral fellow, The Rockefeller University, New York, NY.

Honors

2023	Distinguished Professorship, Duke University
2023	Biophysical Society New and Notable Symposium Lecturer
2022	Duke Science and Technology (DST) scholar
2018	Outstanding Postdoc Mentor Award at Duke University
2018	Hanseong Science Award, Hanseong Sonjaehan Foundation, Korea
2018	The SER-CAT Outstanding Science Award
2018	Biophysical Society New and Notable Symposium Lecturer
2016	NIH Research Program Award, Neuroscience and Disorders of the Nervous System
2016	Biophysical Society New and Notable Symposium Lecturer
2014	NIH EUREKA award, Neuroscience and Disorders of the Nervous System
2012	Biophysical Society New and Notable Symposium Lecturer
2012	NIGMS award, 56th Biophysical Society Annual Meeting
2011-2016	NIH Director's New Innovator Award
2011-2013	Alfred P. Sloan Research Fellow, Alfred P. Sloan Foundation
2011-2013	Basil O'Connor Starter Scholar Research Award, March of Dimes Foundation
2010-2013	Mallinckrodt Scholar, Edward Mallinckrodt, Jr, Foundation
2010-2013	Klingenstein Fellowship Award in the Neurosciences, The Klingenstein Fund
2010-2013	McKnight Scholar Award, The McKnight Endowment Fund for Neuroscience
2009	Whitehead Scholar, Duke University
2004-2007	Postdoctoral Fellow, Jane Coffin Childs Memorial Fund
1998-2003	Predoctoral Fellow, Korean Foundation for Advanced Study

C. Contributions to Science

1. Structure, function, and pharmacology of calcium-permeable Ion channels

Transient receptor potential (TRP) channels are polymodal sensors involved in sensory transduction such as pain and itch. By sensing changes in temperature, ligands (e.g. irritants), and lipids, TRP channels regulate Ca^{2+} flow into neuronal (and non-neuronal) cells. Despite the crucial importance of TRP channels in human physiology and diseases, our understanding of these channels is still far from satisfactory. We have contributed to understanding the mechanisms for sensation of cooling and noxious chemicals by the transient receptor potential (TRP) calcium-permeable channels (TRPM8, TRPV2, TRPV3, TRPA1, TRPML3, and TRPM2) and their regulation by PIP_2. Notably, we have uncovered the molecular basis of heat sensing by the heat and capsaicin sensor TRPV1, the molecular basis of cooling agent sensing by the cold/menthol receptor TRPM8, the mechanism of irritant sensing by the wasabi receptor TRPA1, the mechanism of ligand-dependent gating of TRPV2 and TRPV3. Our work has allowed us to propose fundamental design principles of TRP channels, including the unusual π-helix, reduced symmetry in channel gating and the role of the distal C-terminal domain in TRPV channel regulation.

a) Lejla Zubcevic, Mark A Herzik Jr, Ben C Chung, Zhirui Liu, Gabriel C Lander*, **Seok-Yong Lee***, Cryo-electron microscopy structure of the TRPV2 ion channel. *Nat. Struct. Mol. Biol.*, 2016 Epub Jan 18. doi: 10.1038/nsmb.3159. PMCID: PMC4876856
b) Ying Yin, Son C. Lee, Allen H. Hsu, Mario J. Borgnia, Huanghe Yang, **Seok-Yong Lee***, Structural basis of cooling agent and lipid sensing by the cold activated TRPM8 channel. *Science*, 2019. 363:aav9334. PMCID: PMC6478609
c) Dohoon Kwon, Feng Zhang, Justing G. Fedor, Yang Suo, **Seok-Yong Lee***. "Vanilloid-dependent conformational trajectory of TRPV1 opening revealed through cryoEM ensembles". *Nat. Commun*. 2022. May 24;13(1):2874. PMICID:PMC9130279
d) Ying Yin, Feng Zhang, Shasha Feng, Kevin John Butay, Mario J. Borgnia, Wonpil Im, **Seok-Yong Lee***, Activation mechanism of the mouse cold-sensing TRPM8 channel by cooling agonists and PIP_2. *Science*, 2022. 378, add1268. PMCID: PMC9795508

***** Corresponding author

2. Structure, function, and chemical biology of drug/metabolite transporters

We are interested in a detailed mechanistic understanding of the cellular uptake of drugs and metabolites/nutrients by solute carriers (SLCs), as well as the often-inevitable drug-drug and drug-nutrient interactions that occur. Specifically, we are interested in nucleoside and nucleoside-derived drug transport mediated by Concentrative and Equilibrative Nucleoside Transport proteins (CNTs/ENTs), folate and anti-folate drug transport by the Reduced Folate Carrier (RFC), and organic cation transport by organic cation transporters (OCTs). These transporters are vital to a wide variety of physiological processes including metabolism, cellular signaling, and drug uptake and excretion.

We have elucidated the mechanisms of nucleoside and nucleoside-derived drug uptake by CNT and ENT, two types of nucleoside transporters in humans, and the mechanism of human ENT1 inhibition by adenosine reuptake inhibitor (AdoRI) drugs. From these studies, we have advanced our understanding of the transport model, provided a proof-of-concept to engineer drugs with enhanced selectivity, and provided a platform to develop novel AdoRIs. Our study of RFC and OCTs paves the way for a comprehensive understanding of drug recognition and selectivity by hRFC and OCTs.

a) Marscha Hirschi, Zachary Johnson and **Seok-Yong Lee***, Visualizing multistep elevator-like transitions of a nucleoside transporter. *Nature*, 2017. 545:66-70. PMCID: PMC5567992
b) Nicholas Wright and **Seok-Yong Lee***, Structures of human ENT1 in complex with adenosine reuptake inhibitors, *Nat. Struct. & Mol. Biol*., 2019. 26:599-606. PMCID: PMC6705415
c) Yang Suo, Nicholas Wright, Hugo Guterres, Justin Fedor, Kevin John Butay, Mario J. Borgnia, Wonpil Im, **Seok-Yong Lee***. Molecular basis of polyspecific drug and xenobiotic recognition by OCT1 and OCT2. *Nat. Struct. Mol. Biol.*, 2023 Jul;30(7):1001-1011. PMID:37291422
d) Nicholas Wright, Feng Zhang, Yang Suo, Linyang Kong, Ying Yin, Kedar Sharma, Mario Borgnia, Wonpil Im, **Seok-Yong Lee***. Antiviral drug recognition and elevator-type transporter motions of CNT3. *Nat. Chem. Biol.* 2024 In press, doi:10.1038/s41589-024-01559-8

***** Corresponding author

3. Structural biology of lipid transport in bacterial cell wall synthesis

My recent research has focused on a class of membrane proteins responsible for lipid transport in bacterial cell wall synthesis. Lipid transport in bacterial cell wall synthesis involves Lipid I production and Lipid II flipping, which are carried out by MraY and MurJ, respectively. Both Lipid I production and Lipid II flipping are essential steps in bacterial cell wall synthesis. MraY (phospho-MurNAc-pentapeptide translocase) is considered a very promising target for the development of new antibiotics, as MraY is the target of five different classes of natural product antibiotics and a bacteriolytic protein from bacteriophage phiX174. Significant progress in the field depends upon the structure determination and subsequent mechanistic understanding of MraY and MurJ. We have solved the structure of $MraY_{AA}$, the first structure of a member of the MraY family. We also solved the structures of MraY bound to five different types of peptidyl nucleoside inhibitors, thereby providing the chemical logic of MraY inhibition by naturally occurring nucleoside inhibitors. We have uncovered not only the mechanisms of LLO translocation and flipping by MraY and MurJ in bacterial cell wall synthesis, but also the inhibition of MraY by antibiotics. Our work guides novel approaches to selectively target bacterial cell wall synthesis for antibiotic development. We have recently expanded our program to fungal cell wall synthesis.

a) Ben C. Chung, Ellene H. Mashalidis, Tetsuya Tanino, Mijung Kim, Akira Matsuda, Jiyong Hong, Satoshi Ichikawa, **Seok-Yong Lee***, Structural insights into inhibition of lipid I production in bacterial cell wall synthesis. *Nature,* 2016. 533:557-560. PMCID: PMC4882255
b) Alvin C. Y. Kuk, Ellene H. Mashalidis, **Seok-Yong Lee**[*], Crystal structure of the MOP flippase MurJ in an inward-facing conformation. *Nat. Struc. & Mol. Biol.,* 2016. 24:171-176. PMCID: PMC5382020
c) Alvin C. Y. Kuk, Aili Hao, Ziqiang Guan, **Seok-Yong Lee**[*], Visualizing conformation transitions of the Lipid II flippase MurJ. *Nat. Commun.* 2019. 10:1736. PMCID: PMC6465408
d) Zhenning Ren[‡], Abhishek Chhetri[‡], Ziqiang Guan, Yang Suo, Kenichi Yokoyama*, **Seok-Yong Lee***. "Structural basis for inhibition and regulation of a chitin synthase from *Candida albicans"*. *Nat. Struct. & Mol. Biol*., 2022. Jul;29(7):653-664. PMCID: PMC9359617

***** Corresponding author

Complete List of Published Work in MyBibliography:
https://www.ncbi.nlm.nih.gov/sites/myncbi/seok-yong.lee.1/bibliography/43450645/public/

BIOGRAPHICAL SKETCH

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NAME: Fedor, Justin Gerald

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

POSITION TITLE: Postdoctoral Associate

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

INSTITUTION AND LOCATION	DEGREE (if applicable)	START DATE MM/YYYY	COMPLETION DATE MM/YYYY	FIELD OF STUDY
University of Alberta, Edmonton	BS	09/2006	06/2009	Biochemistry
University of Alberta, Edmonton, Canada	PHD		06/2016	N/A
University of Cambridge, Cambridge	Postdoctoral Fellow	01/2016	12/2019	Biochemistry and Bioenergetics
Duke University, Durham, North Carolina	Postdoctoral Fellow	01/2020	present	Structural Biology and Biochemistry

A. Personal Statement

My Research interests lie in understanding the molecular machinery at the membrane that runs our cells: enzymes and transporters. I seek a mechanistic understanding of how oxidoreductase enzymes facilitate electron transfer and transform their substrates, how transporters selectively bind their substrates/drugs and the molecular motions involved in their activity, and how both these types of proteins engage with the lipids and other proteins in the membrane environment. I have worked on several oxidoreductase systems, from the rather tiny alternative oxidase of Trypanosomes, to E. coli Nitrate Reductase A, and the massive respiratory enzymes and complexes of mitochondria (complex I and its associated supercomplexes). Understanding these systems not only helps us to understand the basic mechanisms of biology, but helps reveal the the causes behind metabolic diseases and aging. More recently, my work has grown to include membrane transporters, namely the reduced folate carrier (SLC19A1) and several related proteins. Studies into these proteins reveals the mechanisms for cellular uptake and distribution of nutrients as well as drugs. Through detailed molecular understanding of these transporters better therapeutic agents can be designed to help with a variety of human diseases, from cancer to inflammation, and mitigate drug-drug and drug-nutrient interactions.

1. Wright NJ, Fedor JG, Zhang H, Jeong P, Suo Y, Yoo J, Hong J, Im W, Lee SY. Methotrexate recognition by the human reduced folate carrier SLC19A1. Nature. 2022 Sep;609(7929):1056-1062. PubMed Central PMCID: PMC9822521.
2. Bridges HR, Fedor JG, Blaza JN, Di Luca A, Jussupow A, Jarman OD, Wright JJ, Agip AA, Gamiz-Hernandez AP, Roessler MM, Kaila VRI, Hirst J. Structure of inhibitor-bound mammalian complex I. Nat Commun. 2020 Oct 16;11(1):5261. PubMed Central PMCID: PMC7567858.
3. Fedor JG, Hirst J. Mitochondrial Supercomplexes Do Not Enhance Catalysis by Quinone Channeling. Cell Metab. 2018 Sep 4;28(3):525-531.e4. PubMed Central PMCID: PMC6125145.
4. Fedor JG, Jones AJY, Di Luca A, Kaila VRI, Hirst J. Correlating kinetic and structural data on ubiquinone binding and reduction by respiratory complex I. Proc Natl Acad Sci U S A. 2017 Nov 28;114(48):12737-12742. PubMed Central PMCID: PMC5715780.

B. Positions and Honors

Positions and Scientific Appointments

2020 - Postdoctoral Associate, Duke University
2016 - 2019 Postdoctoral Researcher, University of Cambridge, Cambridge

Honors

2012 - 2015	Alberta Innovates Health Solutions Graduate Studentship, Alberta Innovates
2010 - 2012	Izaak Walton Killam Memorial Scholarship, University of Alberta
2007 - 2009	Jason Lang Memorial Scholarship, University of Alberta
2017	Medical Research Council End-of-Year Award, Medical Research Council
2016	Madsen Thesis Book Prize, University of Alberta
2010	Faculty of Dentistry and Medicine 75th Anniversary Scholarship, University of Alberta
2009	L.B. Pett Graduate Scholarship in Biochemistry, University of Alberta
2009	Bill Paranchych Memorial Scholarship, University of Alberta
2008	University of Alberta Undergraduate Academic Scholarship, University of Alberta
2006	Canadian Petroleum Producers Award, Canadian Association of Petroleum Producers
2006	Grant MacEwan Arts and Science Faculty Scholarship, Grant MacEwan College
2005	Governor General of Canada Academic Medal, Government of Canada

C. Contribution to Science

1. Electron transfer and catalysis by Nitrate Reductase A

   E.coli is a facultative anaerobe with its ability to flourish under a variety of conditions conferred by its highly dynamic and diverse respiratory enzyme ensemble. Particularly critical is Nitrate Reductase, which allows E. coli to respire on nitrate. This enzyme is a heteromeric oxidoreductase complex comprising a variety of redox cofactors (FeS clusters, hemes and molybdopterin). How the enzyme binds and oxidizes the quinol substrate in the membrane and couples that to the reduction of nitrate at the end of a long electron transport relay can help us understand more sophisticated systems, like the respiratory enzymes in human mitochondria. Using Electron Paramagnetic Resonance spectroscopy and a number of enzymological assays and techniques I biophysically characterized electron transfer between the two hemes within the membrane subunit and probed their role in quinol oxidation. I was able to show how these entities electrostatically influence each other to affect the enzyme activity and proposed a model of quinol oxidation.

   a. Fedor JG, Rothery RA, Weiner JH. A new paradigm for electron transfer through Escherichia coli nitrate reductase A. Biochemistry. 2014 Jul 22;53(28):4549-56. PubMed PMID: 24960296.
   b. Fedor JG, Rothery RA, Giraldi KS, Weiner JH. Q-site occupancy defines heme heterogeneity in Escherichia coli nitrate reductase A (NarGHI). Biochemistry. 2014 Mar 25;53(11):1733-41. PubMed Central PMCID: PMC4059744.

2. Mitochondria are central to many cellular processes, not least of which is the metabolic processes that power the cell. Oxidative phosphorylation is the process by which the energy derived from the food we eat is ultimately converted into ATP, and central to this process is the mitochondrial respiratory complex I. This massive 1 MDa complex of >40 subunits couples the oxidation of NADH to the reduction of membrane-soluble quinone. Quinone is a freely diffusing electron donor within the mitochondrial inner membrane, which in turn is also utilized by complexes II and III. During these electron transfer events, complex I couples this energy to the pumping of protons across the membrane to charge the mitochondria for ATP production by the ATP synthase complex. How complex I accomplishes this exquisitely efficient coupling of electron and proton movements is a very difficult and still poorly understood process. Furthermore, the respiratory enzymes exist as defined supercomplexes of various forms. Various reasons for these supercomplexes have been proposed, but one was that they facilitate quinone channeling between the respiratory enzymes to reduced deleterious side-reactions and ensure efficient electron transfer. My work probed both of these aspects of complex I. Together with my colleagues, we use cryoEM and enzymology techniques to further detailed how quinone interacts with mammalian complex I and proposed a role for the long cavity within which quinone binds in tuning the enzyme kinetics. We then showed how this binding cavity also serves as the binding site of an important respiratory inhibitor, piericidin. Finally, I addressed the hypothesis that mitochondrial supercomplexes channel quinone through an simple and elegant series of experiments that revealed that quinone is not channeled between the respiratory enzymes. These studies

provide further insight into mitochondrial function with implications on ageing and metabolic disease research.

a. Bridges HR, Fedor JG, Blaza JN, Di Luca A, Jussupow A, Jarman OD, Wright JJ, Agip AA, Gamiz-Hernandez AP, Roessler MM, Kaila VRI, Hirst J. Structure of inhibitor-bound mammalian complex I. Nat Commun. 2020 Oct 16;11(1):5261. PubMed Central PMCID: PMC7567858.
b. Fedor JG, Hirst J. Mitochondrial Supercomplexes Do Not Enhance Catalysis by Quinone Channeling. Cell Metab. 2018 Sep 4;28(3):525-531.e4. PubMed Central PMCID: PMC6125145.
c. Fedor JG, Jones AJY, Di Luca A, Kaila VRI, Hirst J. Correlating kinetic and structural data on ubiquinone binding and reduction by respiratory complex I. Proc Natl Acad Sci U S A. 2017 Nov 28;114(48):12737-12742. PubMed Central PMCID: PMC5715780.

3. Folates are essential nutrients for humans, central in the 1- and 2-carbon metabolism of amino acids and DNA. They are taken up by three different systems, with most folates entering the cell via the reduced folate carrier (RFC). Since rapidly dividing cancer cells have higher folate demands many chemotherapeutics are anti-folates that take advantage of RFC or the other systems for entry into cancer cells (proton coupled folate transporter and folate receptors). Knowing how RFC interacts with folate substrates during transport and anti-folate drugs can help design next generation anti-folate drugs. Additionally, comparing the interaction of these drugs with RFC versus the other folate uptake systems can aid in tuning the specificity of drug uptake to limit drug side effects, but a structure of drug-bound RFC was unknown. Using cryoEM, we determined the structure of RFC with the classic chemotherapeutic, methotrexate. This compound does not bind very tightly to RFC as it is a substrate, so to determine its structure we used a chemically modified version of methotrexate which covalently attaches to a single cavity-lining lysine residue, ensure high occupancy of methotrexate in the binding site. Our structural analysis and functional studies identified key residues that stabilize methotrexate binding and substrate exchange, and allowed us to propose a model for drug specificity to RFC.

   a. Wright NJ, Fedor JG, Zhang H, Jeong P, Suo Y, Yoo J, Hong J, Im W, Lee SY. Methotrexate recognition by the human reduced folate carrier SLC19A1. Nature. 2022 Sep;609(7929):1056-1062. PubMed Central PMCID: PMC9822521.

Complete List of Published Work in My Bibliography:
https://www.ncbi.nlm.nih.gov/myncbi/justin.fedor.1/bibliography/public/

D. Scholastic Performance

Scholastic Performance

YEAR	COURSE TITLE	GRADE
	UNIVERSITY OF ALBERTA	
	UNIVERSITY OF ALBERTA, EDMONTON, CANADA	