BIOGRAPHICAL SKETCH

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NAME: Zhou, Ming

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

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
Fudan University, Shanghai, China	B.S.	07/1990	Biochemistry
State University of New York at Buffalo, NY	Ph.D.	07/1999	Biophysics
Rockefeller University, New York City, NY	Postdoc	07/2004	Structural Biology/Biophysics

A. Personal Statement

Research in my lab focuses on structure and function of membrane-embedded enzymes and transporters with the goal of visualizing these proteins in their different functional states and elucidating their mechanisms in terms of the physical and chemical basis of their functions. A project often starts with biochemical characterization and structure determination of an enzyme or a transporter, and progresses to rigorously testing structure inspired hypotheses on mechanisms of the target protein, with a combined approach of structure determination, functional characterizations and molecular dynamics simulations. In addition to advancing basic knowledge, our work also has clinical relevance because the target enzymes and transporters are validated therapeutic targets owing to their roles in well-defined physiological or pathophysiological processes. For structure determination, I made the transition from X-ray crystallography to cryo-electron microscopy (cryo-EM) after joining Baylor College of Medicine (BCM) in 2012. I learnt and practiced cryo-EM procedures at the EM facilities in BCM and Princeton University. These efforts led to structure determination of an ATP-gated ion channel (PMID 31992706), the human diacylglycerol acyltransferase-1 (PMID 32433610), and a mammalian iron transporter ferroportin (PMID 33173040), and established cryo-EM as the main approach for structure determination in my lab.

a. Cao Y., Pan Y., Huang H., Jin X., Levin EJ, Kloss B., and **Zhou M.** (2013). Gating of the TrkH ion channel by its associated RCK protein, TrkA. ***Nature;*** 496 (7445):317-321. PMC3726529
b. Zhou X., Levin E.J., Pan Y., McCoy, J.G., Sharma R., Kloss B., Bruni R., Quick M., **Zhou M.** (2014). Structural basis of the alternating-access mechanism in a bile acid transporter. ***Nature****;* Vol 505: 569-573. PMC4142352
c. Bai, Y., McCoy, J.G., Levin, E.J., Sobrado, P., Rajashankar, K.R., Fox, B.G., **Zhou M.** (2015) X-ray structure of a mammalian stearoyl-CoA desaturase. ***Nature;*** Vol 524: 252-256. PMC4689147
d. Wang, L., Qian, H., Nian, Y., Han, Y., Ren, Z., Zhang, H., Hu, L., Prasad, BVV, Laganowsky, A., Yan, N., **Zhou M.** (2020) Structure and mechanism of human diacylglycerol O-acyltransferase 1. ***Nature***, Vol 581:329-332. PMC7255049

B. Positions and Honors

Positions and Employment

2004 – 2010: Assistant Professor, Department of Physiology & Cellular Biophysics, Columbia University, New York, NY
2010 – 2012: Associate Professor (with tenure), Department of Physiology & Cellular Biophysics, Columbia University, New York, NY

2012 - 2015: Associate Professor (with tenure), Ruth McLean Bowman Bower Endowed Chair, Department of Biochemistry and Molecular Biology, Baylor College of Medicine, Houston, TX

2014 – 2018: Senior Investigator, Kunming Institute of Zoology, Kunming, China. This was an honorary position for teaching and mentoring graduate students. The position does not require FTE commitment.

2015 - : Professor (with tenure), Ruth McLean Bowman Bower Endowed Chair, Department of Biochemistry and Molecular Biology, Baylor College of Medicine, Houston, TX

Other Experience and Professional Memberships

2006-09 Member, American Heart Association Cardiac Electrophysiology Peer Review Study Group
2010-14 Member, Swiss National Science Foundation, Danish Council for Independent Research
2014-17 Member, Proposal Review Panel at SLAC National Accelerator Laboratory
2015-19 Member, NIH BBM study section

Honors

2006 Pew Scholar in Biomedical Science
2006 Alfred P. Sloan Fellow
2006 Basil O'Connor Starter Scholar Award

C. Contributions to Science

My work as an independent investigator has centered on understanding basic mechanisms of membrane proteins. I initially worked on ion channels and gradually expanded into solute transporters and membrane-embedded enzymes.

1 **Regulation of Kv1 channel by cellular redox state.** The Kv1 family of voltage-dependent K^+ channels are important for controlling the timing and frequency of action potentials. Kv1 channels assemble with a cytosolic beta subunit (Kvβ) to form a stable complex. Kvβ is homologous to aldo-keto reductases but it was not clear if Kvβ has enzymatic activity and if so, whether the enzymatic activity has an effect on channel activity. I showed that Kvβ is a functional oxidoreductase and that oxidation of the Kvβ-bound NADPH by either a substrate or an oxidant, such as H_2O_2, increases channel current. My lab then showed that the increase of channel current is due to loss of the N-type inactivation, and that Kvβ interacts with the inactivation gate in a redox dependent manner. My also discovered that cortisone and fluticasone bind to Kvβ and promotes dissociation of an otherwise stable Kv1-Kvβ complex. These studies established the mechanism of redox regulation of Kv1 channel activity and a bridge between cellular redox state and its membrane potential.

a. Weng J, Cao Y, Moss N, **Zhou M.** (2006). Modulation of voltage-dependent *Shaker* family K channel by an aldo-keto reductase. ***Journal of Biological Chemistry****;* Vol 281 (22): 15194-15200. PMCID: PMC2862575.
b. Pan Y, Weng J, Cao Y, Bhosle R, **Zhou M.** (2008). Functional coupling between the Kv1.1 channel and an aldo-keto reductase Kvbeta1. ***Journal of Biological Chemistry****;* Vol. 283 (13): 8634-8642. PMCID: PMC2417172.
c. Pan Y Weng J, Kabaleeswaran V, Li H, Cao Y, Bhosle R, **Zhou M.** (2008). Cortisone dissociates *Shaker* family K^+ channels from their β subunits. ***Nature Chemical Biology***; Vol 4 (11): 708-714. PMCID: PMC2633621.
d. Pan Y, Weng J Levin EJ**, Zhou M.** (2011). Oxidation of NADPH on Kvβ1 inhibits the ball-and-chain type inactivation by restraining the chain. ***Proc. Natl. Acad. Sci. USA***.; Vol. 108 (14): 5885-5890. PMCID: PMC3078402.

2 **Structure and mechanism of PEP group translocation.** The phosphoenolpyruvate-dependent phosphotransferase system (PTS or PEP group translocation) is a sugar uptake system in bacteria. Concentrative sugar uptake is achieved when the incoming sugar is phosphorylated before releasing into the cytosol. Because the energy stored in the phosphoester bond of the phosphorylated sugar is partially recovered when the sugar is metabolized, the PTS system is more energy efficient than the ABC type sugar transporters in which ATP is consumed for sugar uptake. PTS has multiple components and the membrane embedded EIIC component is responsible for sugar transport through cell membrane and was

the only component without a structure. My lab determined crystal structures of EIICs from two different bacterial species. The two structures allowed us to propose structural changes required for substrate translocation, and we were able to test the hypothesis by crosslinking a pair of cysteines strategically placed on one of the EIICs and solving the structure of the crosslinked EIIC. The results led to better understanding of the conformational changes during sugar transport. We also applied single-molecule fluorescence resonance energy transfer (smFRET) and molecular dynamics simulations to validate the structures and to elucidate the dynamics of the EIIC proteins. These results also build a foundation for understanding the mechanism of sugar phosphorylation, which occurs when the sugar remains bound to EIIC to accept a phosphate from another protein, EIIB. We will solve the structure of EIIB-EIIC complex to visualize the last step of sugar uptake.

a. Cao, Y., Jin, X., Levin, E.J., Huang, H., Zong, Y., Quick, M., Weng, J., Pan, Y., Love, J., Punta, M., Rost, B., Hendrickson, W., Javitch, J., Rajashankar, K., & **Zhou M.** (2011). Crystal structure of a phosphorylation-coupled saccharide transporter. ***Nature;*** Vol 473 (7345): 50-54. PMC3201810.
b. McCoy JG, Ren Z, Stanevich V., Lee J., Mitra S., Levin EJ, Poget S., Quick M., Im W., **Zhou M.** (2016) The structure of a sugar transporter of the glucose EIIC superfamily provides insight into the elevator mechanism of membrane transport. ***Structure;*** Vol 24(6):956-64. PMC4899283.
c. Lee J, Ren Z, **Zhou M**, Im W (2017) Molecular Simulation and Biochemical Studies Support an Elevator-type Transport Mechanism in EIIC. ***Biophys J.;*** 112(11):2249-2252. PMCID: PMC5474738.
d. Ren Z, Lee J, Moosa MM, Nian Y, Hu L, Xu Z, McCoy JG, Ferreon ACM, Im W, **Zhou M.** (2018). Structure of an EIIC sugar transporter trapped in an inward-facing conformation. ***Proc Natl Acad Sci U S A.*** Vol 115(23):5962-5967. PMID: 29784777

3 **Structure and mechanism of an ATP-gated ion channel in bacteria.** TrkH is a member of a very large superfamily of putative K^+ transporters (SKT) all of which are composed of four K^+ channel like domains. TrkH assembles with a cytosolic protein TrkA, which is homologous to cytosolic domains found in certain ligand-gated K^+ channels. However, it was not known whether TrkH is a potassium ion channel or whether and how TrkA regulates TrkH activity. My lab expressed and purified TrkH in complex with TrkA and reconstituted the complex into proteoliposomes for patch clamp studies. We recorded single-channel current of TrkH and showed that TrkH is an ion channel that allows permeation of monovalent cations such as Na^+ and K^+ with a slight preference for K^+. We also discovered that ATP increases TrkH open probability through TrkA while ADP inhibits channel opening. We then determined crystal structures of TrkH and TrkH in complex with TrkA in the presence of ATP or ADP. These structures, along with mutational and functional studies, show that binding of ATP induces TrkA to assume a conformation that can open TrkH, and binding of ADP induces a different conformation of TrkA that closes TrkH. These results established coupling between metabolic state of a cell and its membrane potential, and will guide further analysis to understand the function of TrkH-TrkA complex in cells.

a. Cao, Y., Jin, X., Huang, H., Derebe, M., Levin, E.J., Kabaleeswaran, V., Pan, Y., Punta, M., Love, J., Weng, J., Quick, M., Ye, S., Kloss, B., Bruni, R., Martinez-Hackert, E., Hendrickson, W., Rost, B., Javitch, J., Rajashankar, K., Jiang, Y., & **Zhou M.** (2011). Crystal structure of a potassium ion transporter TrkH. ***Nature;*** 471(7338):336-340. PMC3077569.
b. Cao Y., Pan Y., Huang H., Jin X., Levin EJ, Kloss B., and **Zhou M.** (2013). Gating of the TrkH ion channel by its associated RCK protein, TrkA. ***Nature;*** 496 (7445):317-321. PMC3726529
c. Zhang H, Pan Y, Hu L, Hudson MA, Hofstetter KS, Xu Z, Rong M, Wang Z, Prasad BVV, Lockless SW, Chiu W, **Zhou M.** TrkA undergoes a tetramer-to-dimer conversion to open TrkH which enables changes in membrane potential. ***Nature Communications*****.** 2020 Jan 28;11(1):547. PMCID: PMC6987127

4 **Mechanism of urea selectivity and permeation in urea transporters.** Members of the urea transporter family (UT, or SLC1A) mediate rapid and selective transport of urea down its concentration gradient. In mammals, UT is highly expressed in the inner medullary region of the kidney in which urea concentration reaches more than 300 mM. UT is a validated drug target for diuretics because inhibition of UT leads to loss of urea which is the main osmolyte in the kidney. We solved crystal structures of a mammalian UT and a bacterial UT and conducted functional and computational studies. The structures show that UTs transport urea via a channel-like mechanism. The structures also show that UT has a selectivity filter that forms a coin-slot like shape to constrain the orientation of urea and coordinate urea via strategically placed hydrogen donors and acceptors. The structure also suggests a mechanism that prevents water permeation. These results provide insight into the structural basis of urea permeation and selectivity.

a. Levin E.J., Cao Y., Enkavi G., Quick M., Pan Y., Tajkhorshid E., **Zhou M.** (2012). Structure and permeation mechanism of a mammalian urea transporter. ***Proc. Natl. Acad. Sci. USA;*** 109(28):11194-11199. PMC3396522.
b. Levin, E.J., Quick, M., **Zhou, M.** (2009). Crystal structure of a bacterial homologue of the kidney urea transporter. ***Nature;*** 462(7274): 757-761. PMC2871279.

5 **Structure and mechanism of stearoyl-CoA desaturase.** Stearoyl CoA desaturase-1 (SCD1) is a membrane-embedded enzyme that facilitates formation of a double bond to a saturated fatty acid. The reaction is catalyzed by a diiron center and is highly stereo-specific (cis double bond) and regio-specific (9th position). Because desaturation of fatty acid is a crucial step in fat metabolism and in synthesis of cell membranes, SCD1 is a validated drug target for metabolic diseases such as diabetes and for many types of cancers. We solved the structure of mouse SCD1 in complex with its substrate oleoyl-CoA. The structure allows visualization of the active site and reveals how the stereo- and regio-specificities are achieved. However, the diiron center is replaced by two zinc ions in the purified SCD1 and the enzyme is non-functional. The mis-incorporation of zinc ions precluded further mechanistic studies. We overcame this problem by switching to a mammalian cell expression system and obtained fully functional SCD1 properly endowed with iron ions. Because the enzymatic reaction catalyzed by SCD1 requires participation of two additional membrane proteins, cytochrome b and cytochrome b5 reductase, we then developed a method to allow formation of a stable ternary complex of the three proteins for structural and functional studies. These efforts have cleared major barrier for a rigorous study to understand the mechanism of electron transport in SCD1.
a. Bai, Y., McCoy, J.G., Levin, E.J., Sobrado, P., Rajashankar, K.R., Fox, B.G., **Zhou, M.** (2015) X-ray structure of a mammalian stearoyl-CoA desaturase. ***Nature;*** Vol 524: 252-256. PMC4689147
b. Shen J., Wu G., Tsai A., **Zhou M**. (2020) Structure and mechanism of a unique diiron center in mammalian stearoyl-CoA desaturase. ***J Mol Biol.*** Doi:10.1016/j.jmb.2020.05.017 Online ahead of print.

Complete List of Published Work in MyBibliography
https://www.ncbi.nlm.nih.gov/myncbi/ming.zhou.2/bibliography/public/

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

Ongoing Research Support

1R01DK122784 (Multi-PI: Zhou and Tsai) 07/01/2019-05/31/2024
Structure and mechanism of mammalian stearoyl-CoA desaturases
The goal of the project is to reveal the mechanism of substrate recognition and the redox reaction catalyzed by a novel diiron center.

Subaward from R01CA217333 (Nijhawan, Univ. Texas) 04/01/2018-03/31/2023
Tumor-targeted inhibitors of stearoyl-CoA desaturase for the treatment of cancer
The goal of the project is to solve crystal structures of human stearoyl-CoA desaturase in complex with anti-tumor reagents.

Subaward from R01GM132436 (Lockless, Texas A&M) 07/01/2019-06/30/2023
Physiological role for cation channels in bacteria
The goal of the project to express and purify Kch ion channel from E. coli, and to examine its function and structure.