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. Our work also has clinical relevance because many of the proteins are validated therapeutic targets owing to their roles in well-defined physiological or pathophysiological processes. I am proficient with various biophysical and biochemical approaches to investigate mechanisms of enzymes. Although x-ray crystallography had been the main tool for structure determination in my lab, I started to learn cryo-electron microscopy (cryo-EM) after I moved to my current position at Baylor College of Medicine (BCM). I gained extensive hands-on experience at the local cryo-EM facility and at the facility in Princeton University. These efforts have led to structure determination of an ATP-gated ion channel TrkH-TrkA complex (PMID 31992706) and the human diacylglycerol acyltransferase-1 (PMID 32433610). These efforts have also established cryo-EM as the main approach for structure determination in the 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

2015 - : 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.

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 but the function of the beta subunit was not clear. My lab was the first to show that Kvβ is an functional oxidoreductase and we found that oxidation of the Kvβ-bound NADPH significantly increases channel current. We then showed that 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. We also discovered that cortisone and many of its analogs can bind to Kvβ and promotes dissociation of an otherwise very stable Kv1-Kvβ complex. These studies established a pathway by which cellular redox state can regulate 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 unique to bacteria and required 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. We determined crystal structures of two homologous of EIICs, and the two have the same structural fold but in different conformations that illustrate potential structural changes required for substrate translocation. We confirmed the structural changes by crosslinking a pair of cysteines strategically placed on the EIIC, and we obtained structures of both the inward- and outward-facing conformations of the same EIIC. We also conducted single-molecule FRET and molecular dynamics simulations to validate the structural changes. These results also built a foundation for understanding the mechanism of sugar phosphorylation, which occurs when a sugar is translocated to the intracellular side and before it is released into the cytosol. Another

protein, EIIB, transfer a phosphate group to the incoming sugar and we will solve the structure of EIIB-EIIC complex.

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 membrane protein found in many bacteria. TrkH belongs to a very large superfamily of K^+ transporters (SKT) all of which are composed of four homologous domains in a single polypeptide with each domain resembling a simple K^+ channel. TrkH assembles with a cytosolic protein TrkA, which is homologous to cytosolic domains found in certain liganded-gated K^+ channels. However, it was not known whether TrkH is a K^+ channel, and whether TrkA regulates TrkH activity or what ligand TrkA responds to. We 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 showed that ATP binds to TrkA and increases TrkH open probability while ADP inhibits channel opening. We then solved structures of TrkH and TrkH in complex with TrkA in the presence of ATP or ADP with x-ray crystallography and with cryo-EM. These structures, along with mutational and functional studies, show that TrkA-ATP has a conformation that opens TrkH, while TrkA-ADP has a conformation that closes TrkH. These results suggest a 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 SLC14A) 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 channel has a selectivity filter that constrains the orientation of urea and coordinates urea via strategically placed hydrogen donors and acceptors. The structure also suggests a mechanism that prevents water permeation. Our study provides 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 introduces the first 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 (Δ9 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 obesity and diabetes and for many types of cancers. We solved the structure of mouse SCD1 in complex with its substrate oleoyl-CoA. The structure provides visualization of how the stereo- and regio-specificities are achieved, however, it also shows several challenges. The diiron center is replaced by two zinc ions due to the insect cell expression system and as a result, the enzyme is non-functional. In addition, SCD1 requires participation of two additional membrane proteins, cytochrome b_5 and cytochrome b_5 reductase, to complete the reaction cycle. We have developed protocols to produce iron-containing functional SCD1 and we solved structure of SCD1 with a true diiron center to show that the coordination is preserved. We have also expressed and purified full-length cytochrome b_5 and cytochrome b_5 reductase, and we are in the process of assembling all three proteins into a ternary complex for further structural and functional studies.

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 (Zhou and Tsai)
NIH 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 R01GM119396 (Quick, Columbia Univ.) 6/01/2016-03/31/2021
NIH
Molecular mechanism of nucleobase/vitamin C transporters
The goal of the project is to solve structures of an ascorbic acid transporter in different conformations.

Subaward from R01DK061425 (Swaan, Univ. Maryland) 07/01/2016-06/30/2021
Structural biology of the apical bile acid transporter
The goal of the project is to express and purify a eukaryotic homolog of human apical sodium dependent bile acid transporter, and solve its structure by x-ray crystallography.

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.