BIOGRAPHICAL SKETCH

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NAME: cho, uhn-soo

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

POSITION TITLE: Associate 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)	END DATE MM/YYYY	FIELD OF STUDY
Korea University, Seoul	BARCH	02/1998	Agricultural biology
Korea University, Seoul	MS	02/2000	Biochemistry
University of Washington, Seattle, WA	PHD	05/2007	Biological Structure

A. Personal Statement

My research interests focus on elucidating the biological function of physiologically important and structurally challenging proteins or protein complexes based on biochemical and structural observations. I was first introduced to structural biology during college and have since used structural biology as a main tool to answer my scientific questions. As a graduate student in Dr. Wenqing Xu's laboratory at the University of Washington, I pursued the structural and biochemical understanding of protein phosphatase 2A (PP2A), a major Ser/Thr protein phosphatase in eukaryotes. I determined the first crystal structure of a PP2A holoenzyme and provided the structural basis for understanding substrate recognition and complex formation regulated by methylation (Cho & Xu, 2007, Nature; Cho et. al., 2007, PLoS Biology). As a postdoctoral fellow in Dr. Stephen C. Harrison's laboratory at Harvard Medical School, I have focused on elucidating the mechanism of initial kinetochore assembly at centromeres, using structural and biochemical approaches. I have determined crystal structures of two major players in the first stage of kinetochore assembly in budding yeast: the Cse4:H4/Scm3 complex (Cho & Harrison, 2011, PNAS) and Ndc10 (Cho & Harrison, 2011, Nat.Struct.Mol.Biol). As a result of my functional studies, I have outlined a potential sequence of events that occur during initial kinetochore assembly. Currently, I am an associate professor in the Biological Chemistry department at the University of Michigan. As an independent scientist, I have been interested in and focused on four major projects; (1) elucidating the mechanism of epigenetic regulation in eukaryotes, (2) the structural study of methane monooxygenase, (3) understanding the structure-function relationship of human Sestrin2 and its signaling mediators in mTORC1 regulation, and (4) developing a new type of cryo-EM grid that will dramatically improve the sample preparation step in cryo-EM. My contributions to each project led me to publications as a corresponding author in high-impact journals [(1) An et. al., 2015, J.Mol.Biol.; An et al., 2017, eLife.; An et al., 2018, Structure; Park et al., 2019, Nat.Comm.; (2) Lee et. al., 2013, Nature; Kim et. al., 2019, Sci. Adv.; (3) Kim et. al., 2015, Nat. Comm.; (4) Ahn et. al., 2022, JBC; Ahn et. al., 2023, ACS Nano].

Grants;

R01 CA250329 Dou, Cho (MPI) 08/01/2020-05/31/2025 Structural insights into the MLL core complexes

1. Ayoub A, Park SH, Lee YT, Cho US, Dou Y. Regulation of MLL1 Methyltransferase Activity in Two Distinct Nucleosome Binding Modes. Biochemistry. 2022 Jan 4;61(1):1-9. PubMed PMID: 34928138.
2. Lee YT, Ayoub A, Park SH, Sha L, Xu J, Mao F, Zheng W, Zhang Y, Cho US, Dou Y. Mechanism for DPY30 and ASH2L intrinsically disordered regions to modulate the MLL/SET1 activity on chromatin. Nat Commun. 2021 May 19;12(1):2953. PubMed Central PMCID: PMC8134635.

3. Park SH, Ayoub A, Lee YT, Xu J, Kim H, Zheng W, Zhang B, Sha L, An S, Zhang Y, Cianfrocco MA, Su M, Dou Y, Cho US. Cryo-EM structure of the human MLL1 core complex bound to the nucleosome. Nat Commun. 2019 Dec 5;10(1):5540. PubMed Central PMCID: PMC6895043.

B. Positions, Scientific Appointments and Honors

Positions and Scientific Appointments

2019 -	Associate Professor, University of Michigan, Ann Arbor, MI
2012 - 2019	Assistant Professor, University of Michigan, Ann Arbor, MI
2010 - 2013	Special Fellow, Leukemia & Lymphoma Society
2007 - 2012	Postdoctoral Fellow, Harvard Medical School, Boston, MA
2002 - 2007	Ph.D. Graduate Student, Univerisity of Washington, Seattle, WA
2001 - 2002	Postmaster Research Scientist, Los Alamos National Laboratory, Los Alamos, NM
2000 - 2001	Research Scientist, Korea University, Seoul
1998 - 2000	Master Graduate Student, Korea University, Seoul

Honors

2016 - 2017	Junior faculty development award, American Diabetes Association
2010 - 2013	Special Fellow in the Leukemia & Lymphoma Society, the Leukemia & Lymphoma Society
2015	Basil O'Connor Start Scholar Research Award, March of Dimes Birth Defect Foundation
2012	BSSP (Biological Sciences Scholars Program) Scholar Award , University of Michigan

C. Contribution to Science

1. Understanding CENP-A recruiting mechanism at centromeres

   The proper assembly of the kinetochore complex is crucial for the accurate segregation of chromosomes during mitosis. Within this context, CENP-A, a histone H3 variant specific to centromeres, assumes a pivotal role in marking centromeres and recruiting other components essential for kinetochore formation at the centromeric site. Consequently, understanding the specific recruitment of CENP-A into centromeres emerges as a fundamental question that requires attention.

   To unravel this process, we employed structural and biochemical methodologies, investigating it in different organisms, including budding yeast with a point centromere and fission yeast and humans with regional centromeres. Our exploration led us to determine the crystal structures of key components involved in this recruitment process, such as protein phosphatase 2A, shugoshin, the CENP-$A^{Cse4:H4/Scm3}$ complex, and Ndc10. These crystal structures, in conjunction with functional assays, have allowed us to propose a recruitment mechanism for CENP-A^{Cse4} at point centromeres.

   Our primary focus in this research centers on the localization of CENP-A in regional centromeres. For this, we rely on a distinct recruiting factor, the Mis18 holo-complex, responsible for selectively guiding CENP-A to centromeres. Recent progress includes the determination of the crystal structure of Mis16 in complex with histone H4 and Eic1, a component within the Mis18 complex. This has shed light on the centromere-specific role of Mis16 in recognizing CENP-A selectively. Additionally, we have delved into the nuclear translocation mechanism of histone proteins via the karyopherin protein Kap123 in budding yeast.

   Our commitment to understanding the physiological role of the Mis18 holo-complex in CENP-A recruitment remains unwavering. To this end, we employ a multifaceted approach that combines biochemical, structural, and genetic investigations.

   a. An S, Koldewey P, Chik J, Subramanian L, Cho US. Mis16 Switches Function from a Histone H4 Chaperone to a CENP-A(Cnp1)-Specific Assembly Factor through Eic1 Interaction. Structure.

2018 Jul 3;26(7):960-971.e4. PubMed Central PMCID: PMC6031460.

b. An S, Yoon J, Kim H, Song JJ, Cho US. Structure-based nuclear import mechanism of histones H3 and H4 mediated by Kap123. Elife. 2017 Oct 16;6 PubMed Central PMCID: PMC5677370.

c. Cho US, Harrison SC. Ndc10 is a platform for inner kinetochore assembly in budding yeast. Nat Struct Mol Biol. 2011 Dec 4;19(1):48-55. PubMed Central PMCID: PMC3252399.

d. Cho US, Harrison SC. Recognition of the centromere-specific histone Cse4 by the chaperone Scm3. Proc Natl Acad Sci U S A. 2011 Jun 7;108(23):9367-71. PubMed Central PMCID: PMC3111289.

2. Allosteric regulation mechanism of methane monooxygenase (MMO)

The potential of converting methane into methanol offers an environmentally friendly way to harness natural gas as an energy source. However, this chemical transformation requires high temperatures and high pressure, imposing limitations on the use of natural gas as an alternative energy source.

In the methanol production process, methane monooxygenases rely solely on methane as their carbon source. Yet, the intricate chemical mechanism underlying this conversion remains enigmatic, primarily due to a lack of understanding regarding how the regulatory subunits of MMO (Methane Monooxygenase) influence MMO activity.

In a previous study, we unveiled the initial crystal structure of the MMOH-MMOB complex, shedding light on the molecular mechanism through which MMOB enhances the catalytic activity of MMOH. This enhancement occurs by inducing a conformational change in MMOH and its active site, facilitating substrate binding.

Recently, we also determined the crystal structure of the MMOH-MMOD complex, where MMOD acts as an inhibitor. Notably, our findings revealed that MMOD binds to the same binding pocket on MMOH as MMOB. However, MMOD induces distinctive conformational changes in MMOH, leading to the disruption of the active site geometry, in stark contrast to the activation of the di-iron center geometry seen in the case of MMOB. The competition between MMOB and the disruption of the di-iron center geometry is what renders MMOD an inhibitor in this process.

a. Ahn E, Kim B, Park S, Erwin AL, Sung SH, Hovden R, Mosalaganti S, Cho US. Batch Production of High-Quality Graphene Grids for Cryo-EM: Cryo-EM Structure of Methylococcus capsulatus Soluble Methane Monooxygenase Hydroxylase. ACS Nano. 2023 Mar 28;17(6):6011-6022. PubMed Central PMCID: PMC10062032.

b. Kim H, An S, Park YR, Jang H, Yoo H, Park SH, Lee SJ, Cho US. MMOD-induced structural changes of hydroxylase in soluble methane monooxygenase. Sci Adv. 2019 Oct;5(10):eaax0059. PubMed Central PMCID: PMC6774732.

c. Lee SJ, McCormick MS, Lippard SJ, Cho US. Control of substrate access to the active site in methane monooxygenase. Nature. 2013 Feb 21;494(7437):380-4. PubMed Central PMCID: PMC3596810.

3. Structural and functional understanding of human Sestrin2

Sestrins are stress-inducible proteins upregulated by reactive oxygen species and other environmental stresses. Two important physiological functions of Sestrins have been recognized for their roles in antioxidant activity and inhibition of mTORC1 function. Nevertheless, the precise molecular mechanisms governing these functions remain largely unclear. I spearheaded research efforts aimed at uncovering the crystal structure of human Sestrin2. Recently, we successfully determined this structure, offering invaluable insights into how this enzyme effectively carries out its apparently disparate functions.

Our current focus revolves around unraveling the structural intricacies of the GATOR2 complex, a five-component protein assembly responsible for transmitting Sestrin2's inhibitory signals to mTORC1. Given that both the antioxidant and mTORC1 regulation functions of Sestrin are closely linked to aging and diabetes, our investigation holds great promise for shedding light on Sestrin's role in the pathogenesis of human diseases.

a. Kim M, Kowalsky AH, Lee JH. Sestrins in Physiological Stress Responses. Annu Rev Physiol. 2021 Feb 10;83:381-403. PubMed Central PMCID: PMC7878365.
b. Cho CS, Kowalsky AH, Namkoong S, Park SR, Wu S, Kim B, James A, Gu B, Semple IA, Tohamy MA, Solanki S, Cho US, Greenson JK, Shah YM, Kim M, Lee JH. Concurrent activation of growth factor and nutrient arms of mTORC1 induces oxidative liver injury. Cell Discov. 2019;5:60. PubMed Central PMCID: PMC6868011.
c. Ho A, Cho CS, Namkoong S, Cho US, Lee JH. Biochemical Basis of Sestrin Physiological Activities. Trends Biochem Sci. 2016 Jul;41(7):621-632. PubMed Central PMCID: PMC4930368.
d. Kim H, An S, Ro SH, Teixeira F, Park GJ, Kim C, Cho CS, Kim JS, Jakob U, Lee JH, Cho US. Janus-faced Sestrin2 controls ROS and mTOR signalling through two separate functional domains. Nat Commun. 2015 Nov 27;6:10025. PubMed Central PMCID: PMC4674687.

4. Cryo-EM structures of epigenetic regulators in nucleosome recognition

Chromatin modifications play a crucial role in governing the spatial and temporal regulation of gene expression. Many epigenetic regulators target the nucleosome as their substrate to either activate or repress gene transcription epigenetically. Our objective is to unravel the underlying regulatory mechanisms of these epigenetic regulators by elucidating their cryo-EM structures when bound to the nucleosome.

In a recent breakthrough, we successfully determined the cryo-EM structure of human Mixed-Lineage Leukemia (MLL) 1 in complex with the nucleosome. This achievement marks the first depiction of human MLL1 bound to the nucleosome, offering profound insights into how MLL1's methyl-transferase activity can be modulated through its interactions with the nucleosome.

a. Ayoub A, Park SH, Lee YT, Cho US, Dou Y. Regulation of MLL1 Methyltransferase Activity in Two Distinct Nucleosome Binding Modes. Biochemistry. 2022 Jan 4;61(1):1-9. PubMed PMID: 34928138.
b. Lee YT, Ayoub A, Park SH, Sha L, Xu J, Mao F, Zheng W, Zhang Y, Cho US, Dou Y. Mechanism for DPY30 and ASH2L intrinsically disordered regions to modulate the MLL/SET1 activity on chromatin. Nat Commun. 2021 May 19;12(1):2953. PubMed Central PMCID: PMC8134635.
c. Park SH, Ayoub A, Lee YT, Xu J, Kim H, Zheng W, Zhang B, Sha L, An S, Zhang Y, Cianfrocco MA, Su M, Dou Y, Cho US. Cryo-EM structure of the human MLL1 core complex bound to the nucleosome. Nat Commun. 2019 Dec 5;10(1):5540. PubMed Central PMCID: PMC6895043.

5. Development of the affinity grid

Following recent advancements in cryo-electron microscopy (cryo-EM) instrumentation and software algorithms, the next bottleneck in achieving high-resolution cryo-EM structure becomes the sample preparation step. Our objective is to address this challenge by developing a graphene-based affinity cryo-EM grid for the cryo-EM structures of endogenous protein complexes. To achieve this, we will employ a stable cell line expressing the tandem affinity purification (TAP)-tagged target protein and utilize affinity chromatography to purify the target complex. The eluted fraction will contain the ALFA-tagged target protein, which will be concentrated on the graphene-based ALFA-nanobody-attached affinity grid. Following blotting and vitrification, this grid will be used for subsequent structural analysis. Our approach offers solutions to several key challenges in cryo-EM sample preparation, including the reduction in cell quantities required, the direct use of endogenous proteins without overexpression,

and avoiding nonspecific contamination of cell lysates on the affinity cryo-EM grid.

a. Ahn E, Kim B, Park S, Erwin AL, Sung SH, Hovden R, Mosalaganti S, Cho US. Batch Production of High-Quality Graphene Grids for Cryo-EM: Cryo-EM Structure of Methylococcus capsulatus Soluble Methane Monooxygenase Hydroxylase. ACS Nano. 2023 Mar 28;17(6):6011-6022. PubMed Central PMCID: PMC10062032.
b. Ahn E, Tang T, Kim B, Lee HJ, Cho US. Development of an atmospheric plasma jet device for versatile treatment of electron microscope sample grids. J Biol Chem. 2022 Apr;298(4):101793. PubMed Central PMCID: PMC8980800.