BIOGRAPHICAL SKETCH

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NAME: TAYLOR, KENNETH ALLEN

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

POSITION TITLE: DONALD L. D. CASPAR PROFESSOR OF BIOLOGICAL SCIENCE

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
North Carolina State University	BS	05/1969	Textile Chemistry
North Carolina State University	BS	08/1969	Chemistry
North Carolina State University	MS	08/1971	Physical Chemistry
University of California at Berkeley	PHD	05/1975	Biophysics

A. Personal Statement

My laboratory's primary experimental tool is 3-D cryoelectron microscopy (cryoEM). One of our major projects is the 3D imaging of muscle in different structural states. The asynchronous flight muscles of insects are the best ordered muscles in nature and are thus ideal for structural studies. I began my work on the 3-D imaging of insect flight muscle with the late Prof. Michael Reedy (d. 6/18/2019) at the beginning of my first academic appointment at Duke University Medical Center, a collaboration that continued up to his recent death. Before that I started the cryoEM field as a graduate student in Prof. Robert Glaeser's laboratory at U. C. Berkeley. I was the first to demonstrate that molecular images could be obtained from specimens embedded in ice. I then spent almost four years at the MRC Laboratory of Molecular Biology in Cambridge, U.K. where I learned 3-D image reconstruction with the late Dr. L. A. Amos and began my career in muscle research with the late Dr. H. E. Huxley. I continued to work in the muscle field after taking my first academic position at Duke University Medical Center and continued after my relocation to Florida State University, Institute of Molecular Biophysics, in 1995. My research on muscle since my postdoctoral days at the MRC-LMB led to the first 3-D images of the muscle sarcomere in rigor in 1984 using electron crystallography methods on the well-ordered asynchronous flight muscle of *Lethocerus indicus*. We developed Tony Crowther's oblique section reconstruction method to reconstruct several additional biochemical states before adopting electron tomography as our method of choice for muscle imaging. I believe that my research program is the only one that has been able to image myosin head conformations within the muscle lattice during actual contraction using fast frozen, freeze substituted and sectioned muscle. I have been the faculty director of FSU's Biological Science Imaging Resource since 1996, growing it from a facility with a JEOL 1200EX Transmission Electron Microscope (TEM) and a JEOL JSM 840 Scanning Electron Microscope to what it is today with a CM120 BioTwin TEM, recently installed Hitachi HT7800 and a ThermoFisher Titan Krios, the latter with a pair of direct electron detectors, a DE-64 and a K3 mounted on a BioQuantum imaging filter, as well as a Volta Phase Plate. Since 1995, I have mentored 10 PhD students through graduation (I currently have 7), 1 MS student, 14 postdoctoral students, one of which remains with me. One former PhD student is a full professor at Penn State University, the former MS student is a Lecturer at Perimeter College's Alpharetta Campus of Georgia State University, two former postdocs are tenured full professors, one at UCSF, the other at Yale University.

1. Wu S, Liu J, Reedy MC, Perz-Edwards RJ, Tregear RT, Winkler H, Franzini-Armstrong C, Sasaki H, Lucaveche C, Goldman YE, Reedy MK, Taylor KA. Structural changes in isometrically contracting

insect flight muscle trapped following a mechanical perturbation. PLoS One. 2012;7(6):e39422. PubMed PMID: 22761792; PubMed Central PMCID: PMC3382574.

2. Winkler H, Taylor KA. Accurate marker-free alignment with simultaneous geometry determination and reconstruction of tilt series in electron tomography. Ultramicroscopy. 2006 Feb;106(3):240-54. PubMed PMID: 16137829.
3. Taylor KA, Schmitz H, Reedy MC, Goldman YE, Franzini-Armstrong C, Sasaki H, Tregear RT, Poole K, Lucaveche C, Edwards RJ, Chen LF, Winkler H, Reedy MK. Tomographic 3D reconstruction of quick-frozen, Ca2+-activated contracting insect flight muscle. Cell. 1999 Nov 12;99(4):421-31. PubMed PMID: 10571184.
4. Winkler H, Reedy MC, Reedy MK, Tregear R, Taylor KA. Three-dimensional structure of nucleotide-bearing crossbridges in situ: oblique section reconstruction of insect flight muscle in AMPPNP at 23 degrees C. J Mol Biol. 1996 Nov 29;264(2):302-22. PubMed PMID: 8951378.

B. Positions, Scientific Appointments, and Honors

Positions and Employment

1995 -	Professor, Dept. of Biological Science, Florida State University, Tallahassee, FL
1985 - 1995	Research Associate Professor, Dept. of Anatomy, Duke Univ. Medical Center, Durham, NC
1980 - 1985	Research Assistant Professor, Dept. of Anatomy, Duke Univ. Medical Center, Durham, NC
1976 - 1980	Postdoctoral Scientist, MRC Lab. of Molec. Biol., Structural Studies Division, Cambridge, UK
1975 - 1976	Postdoctoral Scientist, University of California, Lawrence Berkeley Laboratory, Berkeley, CA

Other Experience and Professional Memberships

2020 -	Consultant, NIH Common Fund for CryoEM
2018 -	Associate Editor, Journal of Structural Biology
2015 -	Member, American Heart Association
2015 -	Member, American Chemical Society
2014 - 2018	Member, NIGMS Council Advisory Committee on the Structural Biology of AIDS program
2014 -	Member, American Society of Microbiology
2013 - 2013	Member, NIGMS Council Advisory Committee for the PSI:Biology program
2007 - 2011	Member, MI Study Section, later renamed EBIT, NIH Peer Review
1990 -	Editorial Board, Journal of Structural Biology
1980 -	Member, Microscopy Society of America
1980 -	Member, Biophysical Society

Honors

2016	Fellow, Microscopy Society of America
2006	Donald L. D. Caspar Professor of Biological Science, Florida State University
2003	Chair, Gordon Conference on 3-D Electron Microscopy of Macromolecules
2002	Distinguished Research Professor, Florida State University
1994	James A. Shannon Director's Award, NIH
1984	Established Investigator , American Heart Association
1979	Postdoctoral Fellowship, Muscular Dystrophy Fellowship
1978	Postdoctoral Fellowship, NATO
1976	Postdoctoral Fellowship, Jane Coffin Childs Memorial Fund for Medical Research

C. Contributions to Science

1. My first contribution to science was the development of cryoEM which constituted my PhD dissertation research under the direction of Prof. Robert M. Glaeser. For this work, I built (myself) a cold stage, devices for the introduction of prefrozen specimens into the electron microscope and learned how to freeze a thin aqueous film. As we planned to use this method for electron crystallography, at the time single particle image reconstruction being a distant dream, I confined my effort to crystalline specimens from which I was

able to show that the crystallinity of a protein crystal, catalase, was preserved after freezing in liquid nitrogen, and that the contrast was quite good even given the fact that there was no negative stain applied. One particular paper published in the Journal of Microscopy in 1978 was the inspiration for Jacques Dubochet to learn how to freeze thin aqueous specimens into a vitreous ice film from which he was awarded the Nobel Prize. This image showed the kind of detail that a typical frozen-hydrated specimen, not just a crystal, can show in cryoEM. Prof. Robert M. Glaeser and I wrote a retrospective/prospective on this work in 2008, which pointed out the need to prevent protein molecules from aggregating on the air-water interface.

a. Taylor KA, Glaeser RM. Electron diffraction of frozen, hydrated protein crystals. Science. 1974 Dec 13;186(4168):1036-7. PubMed PMID: 4469695.
b. Taylor KA, Glaeser RM. Electron microscopy of frozen hydrated biological specimens. J Ultrastruct Res. 1976 Jun;55(3):448-56. PubMed PMID: 933264.
c. Taylor KA. Structure determination of frozen, hydrated, crystalline biological specimens. J Microsc. 1978 Jan;112(1):115-25. PubMed PMID: 641983.
d. Taylor KA, Glaeser RM. Retrospective on the early development of cryoelectron microscopy of macromolecules and a prospective on opportunities for the future. J Struct Biol. 2008 Sep;163(3):214-23. PubMed PMID: 18606231; PubMed Central PMCID: PMC3291472.

2. For my entire academic career, I have worked on the 3D imaging of the asynchronous flight muscle from the large waterbug *Lethocerus indicus* in a collaboration with the late Mary C. and Michael K. Reedy. The collaboration resulted in the first 3D image of a muscle A-band, published in *Nature* in 1984. Although a major advance, the approach used had major limitations because we could only spatially average only a portion of the unit cell and thus could not directly relate the structure to the X-ray diagram. One solution to this problem was application of Tony Crowther's oblique section reconstruction method, which we developed to its highest level, obtaining 3D images of the entire unit cell, with no missing data. We applied the method to multiple static states and in one publication compared the intensities from the X-ray diagram with the structure factors from the reconstruction. However, this was still not enough because systematic disorder in the filaments disorders the individual myosin heads. The solution to this was electron tomography (ET), which produces a 3D image without averaging but from which subtomograms can be separately aligned and averaged, thus removing the disorder that plagued spatial averaging. We applied ET to several states and for the first time to muscle rapidly frozen while producing active tension which was monitored continuously up to the moment of freezing. This result was published in *Cell* in 1999. Subsequently, we applied multivariate data analysis to individual unit cells, we were able to identify multiple, sometimes completely novel, actin-myosin interactions in a rapidly frozen muscle generating active tension. Finally, our most recent contribution is a 4.2Å structure of the *Lethocerus indicus* myosin tail solved within the native thick filament. This is, I believe, the first and currently only atomic structure of the intact myosin coiled coil domain. It is the longest α-helical coiled coil atomic structure in the PDB.

a. Taylor KA, Schmitz H, Reedy MC, Goldman YE, Franzini-Armstrong C, Sasaki H, Tregear RT, Poole K, Lucaveche C, Edwards RJ, Chen LF, Winkler H, Reedy MK. Tomographic 3D reconstruction of quick-frozen, Ca^{2+}-activated contracting insect flight muscle. Cell. 1999 Nov 12;99(4):421-31. PubMed PMID: 10571184.
b. Wu S, Liu J, Reedy MC, Tregear RT, Winkler H, Franzini-Armstrong C, Sasaki H, Lucaveche C, Goldman YE, Reedy MK, Taylor KA. Electron tomography of cryofixed, isometrically contracting insect flight muscle reveals novel actin-myosin interactions. PLoS One. 2010 Sep 9;5(9)PubMed PMID: 20844746; PubMed Central PMCID: PMC2936580.
c. Hu Z, Taylor DW, Reedy MK, Edwards RJ, Taylor KA. Structure of myosin filaments from relaxed *Lethocerus* flight muscle by cryo-EM at 6 Å resolution. Sci Adv. 2016 Sep;2(9):e1600058. PubMed PMID: 27704041; PubMed Central PMCID: PMC5045269.
d. Rahmani H, Ma W, Hu Z, Daneshparvar N, Taylor DW, McCammon JA, Irving TC, Edwards RJ, Taylor KA. The myosin II coiled-coil domain atomic structure in its native environment. Proc Natl Acad Sci U S A. 2021 April 6;118(14):e2024151118. doi: 10.1073/pnas.2024151118.

3. One of our laboratories signature techniques is "lipid monolayer crystallization", a technique first reported by Roger Kornberg and colleagues. We have gone them one better by publishing six 3D

structures of unstained, frozen hydrated 2-D crystalline arrays. With one of these, we obtained a unique and apparently universal structure for myosin heads in relaxed muscle. Originally found for smooth muscle myosin II, this "relaxed" state is accompanied by instability of the filaments themselves. The asymmetric interaction between myosin heads was subsequently demonstrated by Roger Craig and colleagues for striated muscle. While on a sabbatical with Charles Brooks III, Florence Tama and myself carried out a modelling study which suggested that the formation of the inhibited state of smooth muscle myosin II was accompanied by torsional motions that are propagated through the long α-helical coiled-coil rod domain which forms the thick filament backbone and that these torsional motions might contribute in part to the destabilization of the filaments themselves. We recently obtained experimental evidence for these torsional motions in our structure of the myosin II tail structure, published in PNAS. We obtained a second structure for a myosin inhibited state by combining cryoET with subtomogram averaging of molecular motifs in a 2D array of the cytoplasmic myosin known as myosin V.

a. Wendt T, Taylor D, Trybus KM, Taylor K. Three-dimensional image reconstruction of dephosphorylated smooth muscle heavy meromyosin reveals asymmetry in the interaction between myosin heads and placement of subfragment 2. Proc Natl Acad Sci U S A. 2001 Apr 10;98(8):4361-6. PubMed PMID: 11287639; PubMed Central PMCID: PMC31840.
b. Liu J, Wendt T, Taylor D, Taylor K. Refined model of the 10S conformation of smooth muscle myosin by cryo-electron microscopy 3D image reconstruction. J Mol Biol. 2003 Jun 20;329(5):963-72. PubMed PMID: 12798686.
c. Tama F, Feig M, Liu J, Brooks CL 3rd, Taylor KA. The requirement for mechanical coupling between head and S2 domains in smooth muscle myosin ATPase regulation and its implications for dimeric motor function. J Mol Biol. 2005 Jan 28;345(4):837-54. PubMed PMID: 15588830.
d. Liu J, Taylor DW, Krementsova EB, Trybus KM, Taylor KA. Three-dimensional structure of the myosin V inhibited state by cryoelectron tomography. Nature. 2006 Jul 13;442(7099):208-11. PubMed PMID: 16625208.

4. When I accepted a faculty position at Florida State University, I became a colleague with Kenneth Roux, another electron microscopist and immunologist who wanted to apply his microscopy skill and knowledge of immunology with cryoET to study antibody interactions with the envelope spikes (Env) of HIV and SIV, the first site where antibodies can neutralize the virus. Env consists of two proteins, gp120 and gp41, which are proteolysis products of a larger protein, gp160. gp120 forms the spike arms while gp41 anchors Env to the virus envelope, a lipid bilayer. Our first ET's were done in negative stain and showed the trimeric structure of Env. We later obtained the first 3D images of Env in frozen hydrated virus and proposed a model for the arrangement of the gp41 on the envelope in which the membrane proximal segments extended out from the Env core similar to a tripod.

a. Zhu P, Chertova E, Bess J Jr, Lifson JD, Arthur LO, Liu J, Taylor KA, Roux KH. Electron tomography analysis of envelope glycoprotein trimers on HIV and simian immunodeficiency virus virions. Proc Natl Acad Sci U S A. 2003 Dec 23;100(26):15812-7. PubMed PMID: 14668432; PubMed Central PMCID: PMC307650.
b. Zhu P, Liu J, Bess J Jr, Chertova E, Lifson JD, Grisé H, Ofek GA, Taylor KA, Roux KH. Distribution and three-dimensional structure of AIDS virus envelope spikes. Nature. 2006 Jun 15;441(7095):847-52. PubMed PMID: 16728975.
c. Zhu P, Winkler H, Chertova E, Taylor KA, Roux KH. Cryoelectron tomography of HIV-1 envelope spikes: further evidence for tripod-like legs. PLoS Pathog. 2008 Nov;4(11):e1000203. PubMed PMID: 19008954; PubMed Central PMCID: PMC2577619.
d. Dutta M, Liu J, Roux KH, Taylor KA. Visualization of retroviral envelope spikes in complex with the V3 loop antibody 447-52D on intact viruses by cryo-electron tomography. J Virol. 2014 Nov;88(21):12265-75. PubMed PMID: 25122783; PubMed Central PMCID: PMC4248906.

Complete List of Published Work in MyBibliography (122 items):
https://www.ncbi.nlm.nih.gov/sites/myncbi/kenneth a..taylor.1/bibliography/41086817/public/?sort=date&direction= descending

BIOGRAPHICAL SKETCH

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NAME: Hu, Guiqing

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

POSITION TITLE: Research Faculty I

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
ShanDong Normal University, China, Jinan, ShanDong	BS	1997	Physics
ShanDong Normal University, China, Jinan, Shandong	MS	2000	Physics
Institute of Physics, Chinese Academy of Sciences, Beijing	PHD	2004	Condensed Matter Physics

A. Personal Statement

When I was a graduate student, I used electron microscopy to study the microstructures of some semi-conductor thin films. The graduate training build me strong skill of utilizing the electron microscope, the key equipment for 3D reconstruction, and the ability to handle research projects semi-independently and the ability to summarize the research results in peer-reviewed journals.

Then I joined Brookhaven National Lab as a postdoc. I revealed the structure of the mycobacterium tuberculosis proteasome and mechanism of inhibition by a peptidyl boronate with the hybrid methods of x-ray crystallography and single particle 3D EM. During the postdoc training at BNL, my expertise was expanded with cell culture, protein purification, single particle 3D reconstruction and X-ray crystallography. I was able to read relevant scientific references and devise experiments for a new project and conducted the research project independently and publish the resolved structure.

Then I joined Dr. Taylor's lab at Florida State University. At Taylor Lab, I received the solid training of 3D reconstruction by subtomogram averaging. I have collected cryoEM data and used marker-free tilt series alignment program PROTOMO and the program for sbutomogram averaging I3 to conduct research on various projects such as Insect Flight Muscle, Actin-crosslink rafts, Integrin, HIV/SIV virion with and without antibody and cell migration. The study of the inset flight muscle revealed the contact residues on the myosin II and troponin. Through the actin-crosslink project, a method to reveal atom-atom interaction information from tomogram 3D reconstruction was established. The cross link mechanisms of aldolase and fascin were revealed. For the integrin project, we provided the first proof that talin binding is sufficient to activate and extend membrane-embedded integrin αIIbβ3. we also showed that MnCl2 activated αIIbβ3 integrin alone has a compact conformation, becoming fully upright and open when fibrin bound. For the HIV/SIV project, we showed that the V1/V2 loop located at the top of the virion spike and resolved the controversies about the location of this loop and even the orientation of the atomic structure for the whole spike. For the study of the SIV virion decorated with CD4 and 36D5, We found a multitude of different liganded states of the spikes in a single virus preparation and separated them using statistical analysis. And we also show that CD4 binding and the conformational change in the spikes produced by it enhance the binding of the antibody 36D5. we found the antibody-binding site appears to be the pivot of the motion. For the cell migration project, the tomogram shows the details inside the lamellipodia. The ultra-structure of the lamellipidia and the actin network, the free and ER ribosome and some unknown granular particles inside are revealed. The results of the projects have been published on several peer-reviewed scientific journals and a few manuscripts are also in preparation.

From Oct.2016 to Jan.2017. I worked as a temporary research specialist at Bsir core facility. Responsibilities included coordinating and providing imaging and analytical services to the transmission electron

microscopes(TEMS); routine setting up, alignment and calibration of the TEMS; material ordering; coordinating and assisting instrument maintenance; providing consults and support to users.

Later I have been appointed jointly as postdoctoral associate between Dr. Taylor's lab and Dr. Stagg's lab at Florida State University. For Dr. Taylor's lab, I continue to prepare manuscripts to summarize the results of several projects mentioned above.

At Dr. Stagg's lab, I received training for the Appion pipeline and the automatic data collection on Titan Krios with Leginon and Latitude software. I also received training on image process softwares such as Relion, Cistem, Dynamo and Eman2 tomography. The Appion pipeline was used for both single particle and tomography image processing. Leginon and Latitude are used for automatic data collection for both single particle and tomography projects. Cistem and Relion is used for the single particle reconstruction. And dynamo, I3 and eman2 tomograhpy were used for the tomographic subvolume alignment and classification. At Stagg lab, I have been working on several projects: the 3D structures study of AAV virus bound with AAVR receptor, Clathrin-coated vesicles (CCV), and also Trk-fused gene protein (TFG) . For the Clathrin-coated vesicles (CCV) project, preliminary results were obtained. The tilt series of native CCV extracted from animal organism were collected. The reconstructed tomograms clearly showed the CCV cage, the vesicle underneath the cage and also the substrate inside the vesicle. The results ensure the feasibility of the 3D structure study of the native CCV. For the project of AAV2 bound with AAVR receptor, tilt series were aligned with Protomo and subvolume alignment and classification were conducted with Dyanmo and I3. The results revealed the heterogeneity and the low occupancy of the AAVR receptor. The structure of the AAV2 DJwhole virus at 10 angstrom resolution was obtained. The class averages of AAV2 DJ bound with AAVR at different topology were also obtained. Since full length AAVR receptor shows low occupancy and great topology heterogeneity, truncated AAVR was bound to AAV2 DJ in order to increase the occupancy, decrease the heterogeneity and improved the resolution of the complex structure. Structure of AAV2 DJ bound with truncated AAVR was obtained at 2.4 angstrom resolution. The results of single particle and tomography together revealed the binding mechanism of the AAV2 DJ and AAVR. The information revealed will benefit the design of gene therapy. The results are published on elife. TFG has preferred orientation in vitrified ice and this causes difficulty for single particle 3D reconstruction. Tomography was used to reconstruct the structure of TFG which can be used as initial model for single particle reconstruction. At Stagg lab, my expertise in cryo-electron tomography was strengthened and expanded and my expertise in single particle 3D reconstruction is updated and further extended.

From March 2019 I was appointed as research faculty I at Institute of Molecular Biophysics, Florida State University. At Taylor's lab, outside the projects utilizing tomography and following image process, new project utilizing both single particle 3D reconstruction and cryo-tomography is initiated. The new reconstruction will aim for atomic resolution to resolve the structure of vertebrate thick filament. My extensive skills and experiences in cryo-electron tomography and single particle 3D reconstruction will directly contribute to the new project. At Stagg lab, single particle and tomography are continued to be used to study the binding mechanism of AAVR receptor with native AAV2 and also other AAV family member such as AAV5. Eman2 tomography is utilized for future work on the AAV project together with protomo, i3 and dynamo.

In summary, I have a broad background in structural biology with specific training and expertise which includes cryo-electron tomography, single particle 3D EM, x-ray crystalllogarphy, cell culture and protein purification. I have demonstrated record of successfully and productive research projects and also competency of scientific manuscript preparation. My solid research skill and expertise qualify me to take part in the study of complex structures by cryo-electron tomography, single particle 3D reconstruction and X-ray crystallography.

1. Hu G, Liu J, Taylor KA, Roux KH. Structural comparison of HIV-1 envelope spikes with and without the V1/V2 loop. J Virol. 2011 Mar;85(6):2741-50. PubMed PMID: 21191026; PubMed Central PMCID: PMC3067966.
2. Meyer N, Hu G, Davulcu O, Xie Q, Noble A, Yoshioka C, Gingerich D, Trzynka A, David L, Stagg S, Chapman M. Structure of the gene therapy vector, adeno-associated virus with its cell receptor. eLife. 2019 May 22; 8.
3. Hu G, Taylor DW, Liu J, Taylor KA. Identification of interfaces involved in weak interactions with application to F-actin-aldolase rafts. J Struct Biol. 2018 Mar;201(3):199-209. PubMed PMID: 29146292; NIHMSID: NIHMS922733; PubMed Central PMCID: PMC5820182.

4. Hu G, Liu J, Roux KH, Taylor KA. Structure of Simian Immunodeficiency Virus Envelope Spikes Bound with CD4 and Monoclonal Antibody 36D5. J Virol. 2017 Aug 15;91(16)PubMed PMID: 28539445; PubMed Central PMCID: PMC5533903.

B. Positions and Honors

Positions and Employment

2000 - 2004	PHD candidate, Institute of Physics, Chinese Academy of Sciences, Beijing
2004 - 2006	Research Associate, Brookhaven National Laboratory, Upton, NY
2006 - 2019	Research Associate, Florida State University, Tallahassee, FL
2019 -	Research Faculty I, Florida State Univeristy, Tallahassse, FL

Other Experience and Professional Memberships

Honors

2002	Excellent graduate student award, Inst. of Phys., Chinese Academy
2009	Travel award, GRC on three dimensional electron microscopy
2010	travel award, Recent advances and future prospects for visualizing macromolecular complexes and cellular structures workshop

C. Contribution to Science

1. When I was a graduate student, I used electron microscopy to study the microstructures of semi-conductor thin films such as GaN Films Grown on Si(111) and AlAs/GaAs. I successfully handle the projects (e.g. experiment designation, data collecting, data analysis, presentation and summarization), collaborated with other researchers, and produced several peer-reviewed publications.

    a. Kong,X.,, Hu,G.Q.,, Duan,X.F.,, Lu,Y.,, Liu,X.L.,. Polarity determination for GaN thin films by ELECTRONenergy loss spectroscopy. APPL Phys. Lett.. 2002; 81(11):1990.
    b. Wang,Y.Q.,, Hu,G.Q.,, Duan,X.F.,, Sun,H.L.,, Xue,Q.K.,. Microstructure and formation mechanism of titanium dioxide nanotubes. Chemical Physics Letters. 2002; 365(5-6):427.
    c. Hu,G.Q.,, Wan,L.,, Duan,X.F.,, Chen,H.,, Li,D.S.,, Han,Y.J.,, Huang,Q.,, Zhou,J.M.,. Transmission electron microscopy and atomic force microscopy studies of GaN films grown on AlAs/GaAs(001) substrates. Journal of Crystal Growth. 2003; 252(4):517.
    d. Hu,G.Q,, Wan,L,, Kong,X,, Wang,Y.Q.,, Duan,X.F.,, Lu,Y.,, Liu,X.L.,. Microstructure of GaN Films Grown on Si(111) Substrates by Metalorganic chemical Vapor DEPOSITION. Journal of Crystal Growth. 2003; 256:416.

2. When I was postdoc at Brookhaven National Laboratorr, I revealed the Structure of the Mycobacterium tuberculosis proteasomeand mechanism of inhibition by a peptidyl boronate with the hybrid methods of x-ray crystallography and single particle 3D EM. During the postdoc training at BNL, my expertise was expanded with cell culture, protein purification, Single particle 3D reconstruction and X-ray crystallography. I was able to read relevant scientific references and devise experiments for a new project and conducted the research project independently and publish the resolved structure.

    a. Hu G, Lin G, Wang M, Dick L, Xu RM, Nathan C, Li H. Structure of the Mycobacterium tuberculosis proteasome and mechanism of inhibition by a peptidyl boronate. Mol Microbiol. 2006 Mar;59(5):1417-28. PubMed PMID: 16468986.
    b. Lin G, Hu G, Tsu C, Kunes YZ, Li H, Dick L, Parsons T, Li P, Chen Z, Zwickl P, Weich N, Nathan C. Mycobacterium tuberculosis prcBA genes encode a gated proteasome with broad oligopeptide specificity. Mol Microbiol. 2006 Mar;59(5):1405-16. PubMed PMID: 16468985.

3. 3) For my work at Taylor lab at Florida State University, I received the solid training of 3D reconstruction by subtomogram averaging. I have collected cryoEM data and used well-known marker-free tilt series alignment program PROTOMO and the program for sbutomogram averaging I3 to conduct research on various projects such as Insect Flight Muscle, Actin-crosslink rafts, Integrin, HIV/SIV virion with and without antibody and cell migration. The study of the inset flight muscle revealed the contact residues on the myosin II and troponin. It is the first time that the interaction between myosin II and troponin was revealed at the residue level. Through the actin-crosslink project, a method to reveal atom-atom interaction information from tomogram 3D reconstruction was established. The cross link mechanisms of aldolase and fascin were revealed. For the integrin project, we provided the first proof that talin binding is sufficient to activate and extend membrane-embedded integrin αIIbβ3. we also showed that MnCl2 activated αIIbβ3 integrin alone has a compact conformation, becoming fully upright and open when fibrin bound. For the HIV/SIV project, we showed that the V1/V2 loop located at the top of the virion spike and resolved the controversies about the location of this loop and even the orientation of the atomic structure for the whole spike. For the study of the SIV virion decorated with CD4 and 36D5, We found a multitude of different liganded states of the spikes in a single virus preparation and separated them using statistical analysis. And we also show that CD4 binding and the conformational change in the spikes produced by it enhance the binding of the antibody 36D5. we find the antibody-binding site appears to be the pivot of the motion. For the cell migration project, the tomogram shows the detail inside the lamellipodia. The ultra-structure of the lamellipidia and the actin network, the free and ER ribosome and some unknown granular particles inside are revealed. Outside the projects utilizing tomography and following image process, new projects utilizing single particle 3D reconstruction is initiated. The new single particle 3D reconstruction will aim for atomic resolution to resolved the structure of thick filament. My extensive skills and experiences in cryo-electron tomography and single particle 3D reconstruction will directly contribute to the new project.

    a. Hu G, Taylor DW, Liu J, Taylor KA. Identification of interfaces involved in weak interactions with application to F-actin-aldolase rafts. J Struct Biol. 2018 Mar;201(3):199-209. PubMed PMID: 29146292; PubMed Central PMCID: PMC5820182.
    b. Hu G, Liu J, Roux KH, Taylor KA. Structure of Simian Immunodeficiency Virus Envelope Spikes Bound with CD4 and Monoclonal Antibody 36D5. J Virol. 2017 Aug 15;91(16)PubMed PMID: 28539445; PubMed Central PMCID: PMC5533903.
    c. Hu G, Liu J, Taylor KA, Roux KH. Structural comparison of HIV-1 envelope spikes with and without the V1/V2 loop. J Virol. 2011 Mar;85(6):2741-50. PubMed PMID: 21191026; PubMed Central PMCID: PMC3067966.
    d. Ye F, Hu G, Taylor D, Ratnikov B, Bobkov AA, McLean MA, Sligar SG, Taylor KA, Ginsberg MH. Recreation of the terminal events in physiological integrin activation. J Cell Biol. 2010 Jan 11;188(1):157-73. PubMed PMID: 20048261; PubMed Central PMCID: PMC2812850.

4. 4) At Dr. Stagg's lab, I received training for the Appion pipeline and the automatic data collection on Titan Krios with Leginon and Latitude software. I also received training on image process softwares such as Relion, Cistem, Dynamo and Eman2 tomography. The Appion pipeline was used for both single particle and tomography image processing. Leginon and Latitude are used for automatic data collection for both single particle and tomography projects. Cistem and Relion is used for the single particle reconstruction. And dynamo, I3 and eman2 tomograhpy were used for the tomographic subvolume alignment and classification. At Stagg lab, I have been working on several projects: the 3D structures study of AAV virus bound with AAVR receptor, Clathrin-coated vesicles (CCV), and also Trk-fused gene protein (TFG) . For the Clathrin-coated vesicles (CCV) project, preliminary results were obtained. The tilt series of native CCV extracted from animal organism were collected. The reconstructed tomograms clearly showed the CCV cage, the vesicle underneath the cage and also the substrate inside the vesicle. The results ensure the feasibility of the 3D structure study of the native CCV. For the project of AAV2 bound with AAVR receptor, tilt series were aligned with Protomo and subvolume alignment and classification were conducted with Dyanmo and I3. The results revealed the heterogeneity and the low occupancy of the AAVR receptor. The structure of the AAV2 DJwhole virus at 10 angstrom resolution was obtained. The class averages of AAV2 DJ bound with AAVR at different topology were also obtained. Since full length AAVR receptor shows low occupancy and great topology heterogeneity, truncated AAVR was bound to AAV2 DJ in order to increase the occupancy, decrease the heterogeneity and improved the resolution of the complex structure. Structure of AAV2 DJ bound with truncated AAVR was obtained at 2.4 angstrom resolution. The results of single

particle and tomography together revealed the binding mechanism of the AAV2 DJ and AAVR. The information revealed will benefit the design of gene therapy. The results are published on elife. Single particle and tomography are continued to be used to study the binding mechanism of AAVR receptor with native AAV2 and also other AAV family member such as AAV5. Eman2 tomography is utilized for future work together with protomo, i3 and dynamo. TFG has preferred orientation in vitrified ice and this causes difficulty for single particle 3D reconstruction. Tomography was used to reconstruct the structure of TFG which can be used as initial model for single particle reconstruction. At Stagg lab, my expertise in cryo-electron tomography was strengthened and expanded and my expertise in single particle 3D reconstruction is updated and further extended.

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