BIOGRAPHICAL SKETCH

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NAME: Luger Karolin, Ph.D.

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POSITION TITLE: Professor, Dept. of Chemistry and Biochemistry, and Jennie-Smoly-Caruthers Endowed Chair of Chemistry and Biochemistry, University of Colorado; Investigator, Howard Hughes Medical Institute

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	Completion Date MM/YYYY	FIELD OF STUDY
University of Innsbruck, Austria	B.S.	05/1983	Biology / Microbiology
University of Innsbruck, Austria	M.S.	05/1986	Biology / Biochemistry
University of Basel (Biocenter), Switzerland	Ph.D.	02/1989	Biochemistry/ Biophysics
Swiss Federal Institute of Technology, Switzerland	Postdoc	12/1994	Structural Biology

A. Personal Statement

My lab studies the structure and function of large macromolecular assemblies involved in chromosome organization. We have years of experience in studying the structural biology of nucleosomes and chromatin and associated proteins. More recently, we have investigated the role of the posttranslational modifications of histones on nucleosome and chromatin structure and dynamics, and the structure and mechanism of histone chaperones in nucleosome assembly and disassembly. We are working towards a quantitative description of nucleosome assembly and disassembly, and are investigating the role of histone chaperones in these processes.
Our approaches are mostly quantitative and include x-ray crystallography, analytical ultracentrifugation, fluorescence-based affinity assays, hydrogen-deuterium exchange coupled to mass spectrometry, small angle x-ray scattering, atomic force microscopy, and more recently fluorescence imaging in live cells. My research is highly collaborative. For example, I have been an active participant on a Program Project Grant and other multi-PI grants. I have a long-standing history of working with undergraduates, graduate students and postdocs. Throughout my career, I have served in numerous advisory functions to the NIGMS (e.g. regular and ad hoc member of study sections, site visits, Center Grant reviews, and NAGMS council).

B. Positions and Honors

1990-1994	Postdoctoral Fellow, Dept. of Molecular Biology and Biophysics, Swiss Federal Institute of Technology (ETH), Zürich, Switzerland
1994-1999	Research Assistant Professor (‘Oberassistent’), ETH Zürich, Switzerland
1999-2003	Assistant Professor, Dept. of Biochemistry and Molecular Biology, Colorado State University
2003-2007	Associate Professor, Dept. of Biochemistry and Molecular Biology, Colorado State University
2004-2007	Monfort Professor, Dept. of Biochemistry and Molecular Biology, Colorado State University
2005-present	Adjoint Professor, Dept. of Biochemistry and Molecular Genetics, Colorado School of Medicine
2005-present	Investigator, Howard Hughes Medical Institute
2007-2015	University Distinguished Professor, Colorado State University
2015-present	Affiliate Professor, Dept. of Biochemistry and Molecular Biology, Colorado State University

2015-present Professor, Dept. of Chemistry and Biochemistry, University of Colorado
2015-present Jennie-Smoly-Caruthers Endowed Chair of Chemistry and Biochemistry, University of Colorado

***Honors / Professional Service*:**

1999 Searle Scholar Award
2000 Basil O'Conner Starter Scholar Award
2002 Reviewer, NASA Biomaterials study section
2003 Ad hoc reviewer, NIH BBCB study section
2003 Reviewer, NIH training grants study section
2004 Monfort Professor Award
2005-2009 NIH MSFC study section, regular member
2005-2014 Keystone Symposia, member of the Scientific Advisory Board
2007-present University of Colorado Cancer Center member
2007 Vorarlberg, Austria, State Science Prize
2008-present Journal of Biological Sciences, Editorial Board
2009-2013 EU Marie Curie Nucleosome 4D Network, SAB member and Visiting Scientist
2010 NIH, College of CSR Reviewers
2010-present IRSF, member of the Scientific Review Board
2010-2013 National Advisory General Medical Sciences Council
2011-2012 NIGMS Protein Structure Initiative Advisory Committee, Chair
2012 4DCellFate project SAB member
2013 Biophysical Society National Lecturer
2014 Biophysical Society Fellow
2015 Jack E. Cermak Undergraduate Advising Award
2016 HHMI Faculty Scholars Mentoring Board

C. Contribution to Science

Nucleosome structure

In 1997, the central role of chromatin and nucleosomes in the regulation of transcription, replication, and DNA repair had fully emerged. As a postdoctoral researcher in the lab of Dr. T. Richmond, I was the lead author on a manuscript determining the structure of the nucleosome core particle, the fundamentally repeating unit of chromatin (1). This was widely heralded as a breakthrough discovery (e.g. Science magazine – breakthroughs of the year 1997), and made its way in every biochemistry and cell biology text book. Completion of this project entailed the development of methods to prepare recombinant nucleosomes (2) that are now used in every lab that analyzes nucleosomes in vitro. Upon establishing my own lab, we determined the first structure of a nucleosome in complex with another protein, a peptide from the LANA Protein of Kaposi Sarcoma Herpes Virus (3), and identified the acidic patch on the surface of the nucleosome as a key determinant of nucleosome-nucleosome interactions (4). Subsequently, others showed that the acidic patch is an important docking region for many other nucleosome-interacting proteins. Most recently, we have investigated the structure of archeal chromatin to understand the origins of the eukaryotic nucleosome.

Structure of nucleosomes with post-translational modifications and histone variants

When I started my own lab in 1999, the prevailing hypothesis was that epigenetic modifications of chromatin, such as acetylation or methylation, but also incorporation of histone variants, would structurally alter nucleosome to provide a mechanism for regulating access to DNA. We expanded our studies on nucleosome structure to include nucleosomes containing histone variants (5) (6); histones with post-translational modifications (7), and biologically relevant 'sin' mutants (8). Our studies revealed that these structures were very similar to that of unmodified nucleosomes. We made the important conclusion that crystallization stabilizes nucleosomes, thereby obscuring any changes in their dynamic properties, and this led us to develop

assays to investigate nucleosome stability, dynamics, and the mechanisms of nucleosome assembly. In some cases, post-translational modifications lead to changes in chromatin higher order structure (7).

Nucleosome stability and dynamics

Nucleosomes are multi-component assemblies that undergo a number of structural transitions, as shown in a collaborative studies with two labs specializing in single molecule studies (9, 10). These studies revealed a multitude of finely tuned interactions that hold together the nucleosome, and show evidence for a new dynamic state in which the interface between H2A-H2B dimer and $(H3\text{-}H4)_2$ is exposed. As such, the thermodynamic properties of the nucleosome are difficult to define. We have developed a novel approach to measure nucleosome stability that allows us to thermodynamically define the various steps in nucleosome assembly / disassembly (11), and are in the process of further refining this. These investigations are made possible by our powerful HI-FI assay which we developed to accurately measure protein-protein and protein-DNA affinities in solution and with high reproducibility (12).

Histone chaperone Nap1: structure and function

Our interest in the mechanism of nucleosome assembly and disassembly led us to determine the structure of a conserved assembly factor; Nucleosome Assembly Protein 1 (Nap1). This structure represents a novel fold (13) that is representative of the large Nap1 family, as shown by us for the yeast protein Vps75 (14), and by others. This was followed by a rigorous description of Nap1 interactions with histones (15). We further showed that Nap1 shields histone surfaces used in a nucleosome and can put H2A-H2B in an unconventional tetrameric form (16). This latter investigation made extensive use of hydrogen/deuterium exchange coupled to mass spectrometry (HDX/MS) to characterize protein-protein interfaces, and we will further exploit this approach in the future. We are currently using Nap1 as a tool to study the mechanism of nucleosome assembly in detail.

Other histone chaperones

We also investigate the structure and function of histone chaperones FACT (17) and CAF-1. We have also delved into the mechanism by which centromeric nucleosomes are assembled by the histone chaperones CAL1 (Drosophila; (18)) and Scm3 (yeast; (19)). Most recently, our studies on the interaction of the abundant nuclear enzyme Poly [ADP-ribose] polymerase 1 (PARP-1) have revealed that auto-modification resulting in the attachment of long poly-ADP-ribose chains to itself alters its interactions with chromatin, and confers on PARP-1 the activity of a powerful histone chaperone (20). PARP-1 is a key player in the recognition of DNA damage and initiates DNA damage response; it is also a regulator of transcription. To our knowledge this is the first protein that is switched on by a post-translational modification. Due to the presence of glycohydrolases in the nucleus, PARylation is a very transient modification. Thus, this post-translational modification provides the means for a localized burst of histone chaperone activity. Such a ‘flash mob’ activity makes a lot of sense in both functional contexts.

1. Luger K, Mader AW, Richmond RK, Sargent DF, & Richmond TJ (1997) Crystal structure of the nucleosome core particle at 2.8 A resolution. *Nature* 389(6648):251-260.
2. Dyer PN*, et al.* (2004) Reconstitution of nucleosome core particles from recombinant histones and DNA. *Methods Enzymol* 375:23-44.
3. Barbera AJ*, et al.* (2006) The nucleosomal surface as a docking station for Kaposi’s sarcoma herpesvirus LANA. *Science* 5762:856-861.
4. Chodaparambil JV*, et al.* (2007) A charged and contoured surface on the nucleosome regulates chromatin compaction. *Nat Struct Mol Biol* 14(11):1105-1107.
5. Suto RK, Clarkson MJ, Tremethick DJ, & Luger K (2000) Crystal structure of a nucleosome core particle containing the variant histone H2A.Z. *Nat Struct Biol* 7(12):1121-1124.
6. Chakravarthy S*, et al.* (2005) Structural characterization of the histone variant macroH2A. *Mol Cell Biol* 25(17):7616-7624.
7. Lu X*, et al.* (2008) The effect of H3K79 dimethylation and H4K20 trimethylation on nucleosome and chromatin structure. *Nature Structural & Molecular Biology* 15(10):1122-1124.
8. Muthurajan UM*, et al.* (2004) Crystal structures of histone Sin mutant nucleosomes reveal altered protein-DNA interactions. *Embo J* 23(2):260-271.
9. Bohm V*, et al.* (2011) Nucleosome accessibility governed by the dimer/tetramer interface. *Nucleic Acids Res* 39(8):3093-3102.

10. Sheinin MY, Li M, Soltani M, Luger K, & Wang MD (2013) Torque modulates nucleosome stability and facilitates H2A/H2B dimer loss. *Nat Commun* 4:2579.
11. Andrews AJ, Chen X, Zevin A, Stargell LA, & Luger K (2010) The Histone Chaperone Nap1 Promotes Nucleosome Assembly by Eliminating Nonnucleosomal Histone DNA Interactions. *Mol Cell* 37(6):834-842.
12. Hieb AR, D'Arcy S, Kramer MA, White AE, & Luger K (2012) Fluorescence strategies for high-throughput quantification of protein interactions. *Nucleic Acids Res* 40(5):e33.
13. Park YJ & Luger K (2006) The structure of nucleosome assembly protein 1. *PNAS* 103(5):1248-1253.
14. Park YJ, Sudhoff KB, Andrews AJ, Stargell LA, & Luger K (2008) Histone chaperone specificity in Rtt109 activation. *Nat Struct Mol Biol* 15(9):957-964.
15. Andrews AJ, Downing G, Brown K, Park YJ, & Luger K (2008) A thermodynamic model for Nap1-histone interactions. *J Biol Chem* 283(47):32412-32418.
16. D'Arcy S*, et al.* (2013) Chaperone Nap1 Shields Histone Surfaces Used in a Nucleosome and Can Put H2A-H2B in an Unconventional Tetrameric Form. *Mol Cell* 51(5):662-677.
17. Winkler DD, Muthurajan UM, Hieb AR, & Luger K (2011) Histone chaperone FACT coordinates nucleosome interaction through multiple synergistic binding events. *J Biol Chem* 286(48):41883-41892.
18. Chen CC*, et al.* (2014) CAL1 is the Drosophila CENP-A assembly factor. *J Cell Biol* 204(3):313-329.
19. Dechassa ML, Wyns K, & Luger K (2014) Scm3 deposits a (Cse4-H4)2 tetramer onto DNA through a Cse4-H4 dimer intermediate. *Nucleic Acids Res* 42(9):5532-5542.
20. Muthurajan UM*, et al.* (2014) Automodification switches PARP-1 function from chromatin architectural protein to histone chaperone. *PNAS* 111(35):12752-12757.

http://www.ncbi.nlm.nih.gov/sites/myncbi/karolin.luger.1/bibliography/40851519/public/?sort=date&direction=ascending

D. Research Support

***Ongoing Research Support*:**

Howard Hughes Medical Institute 9/16/2005 – 12/31/2021
Dr. Luger's salary and benefits and those of two postdocs, a lab manager, and admin coordinator, are paid by HHMI, *exclusive of Dr. Luger's salary and benefits*. HHMI provides funds for operations, which includes travel, supplies, equipment maintenance and equipment under $10,000. All HHMI budgets are determined annually.

Completed Research Support:

GM067777 (Luger) 5/1/2003 – 8/31/2016 NIGMS/NIH (**currently in NCE**)
Title: Functional connections between histone variants and histone chaperones
Goal: The machinery that transcribes and replicates the information encoded in the human genome is prevented access due to the packaging of all genomic DNA with an equal amount of protein to form chromatin. Here we study two functionally related activities that regulate DNA accessibility and thus vital cellular processes through the modulation of chromatin structure. We are using quantitative and structural approaches to gain insight into how structural transitions within chromatin allow vital biological processes to occur.

1P01GM088409 (Nyborg) 5/1/2010 – 4/30/2015 NIGMS/NIH
Title: Histone Chaperones and Acetyltransferases in Chromatin Transitions
Goal: A detailed knowledge of the mechanisms that regulate chromatin structure is a prerequisite for understanding how misguided compaction/decompaction of highly condensed genomic DNA may lead to disease through misregulation of gene transcription. This project (R01 scope) addresses the interconnection between two seemingly unrelated activities involved in the modulation of chromatin structure.

GM096863-02S1 (Kutateladze) 8/2/2012 – 4/30/2015 NIGMS/NIH
Title: Molecular analysis of CHD4
Goal: Consortium agreement to determine the structure of PHD12 domain in complex with mononucleosomes higher order chromatin (tri-nucleosomes).

1R01GM096192 (Breidt) 8/20/2010 – 7/31/2014 NIGMS/NIH
Title: The Inverse Problem for Estimation of Structure of Biological Macromolecules from Small-Angle

X-Ray Scattering Data

Goal: A systematic statistical, mathematical, and computational investigation of the inverse problem associated with molecular reconstructions from small angle X-ray scattering data.