BIOGRAPHICAL SKETCH

NAME: Mathias Lösche

POSITION TITLE: Professor of Physics; Professor of Biomedical Engineering (by courtesy); NIST Associate

eRA COMMONS USER NAME: MLoesche

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include post-doctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE	Completion Date	FIELD OF STUDY
University of Ulm, Germany	B.A.	1979	Physics
University of Ulm, Germany	M.A.	01/1983	Physics
Technical University of Munich, Germany	Ph.D.	05/1986	Biophysics
University of California, San Diego, CA	Post-Doc	03/1988	Biophysics

A. Personal Statement

Building upon a traditional Physics curriculum in Germany, I spent the first portion of my career on the physical chemistry of interfaces in the context of materials science, colloids and polymeric surface films. Since transitioning from a tenured faculty position in Germany to the U.S. in 2003, I shifted my research focus to the membrane biophysics. One major tool in this research is neutron scattering, specifically neutron reflection (NR), conducted at the NIST Center for Neutron Research (NCNR) and the Spallation Neutron Source (SNS) at Oak Ridge National Laboratory. This “tip-of-the-iceberg” technology is complemented with various surface-sensitive optical, fluorescence, electrochemical and x-ray based characterization techniques located in our labs at Carnegie Mellon [1]. Here, we developed novel characterization capabilities for proteins associated with disordered, fluid membranes for structural and dynamic investigations. In neutron scattering, our advanced sample preparation and data handling routines enable us to determine the organization of proteins at membrane surfaces with unprecedented detail and precision. No other research group world-wide has such a combination of high-quality instrumentation and experimental know-how at their disposal. To rationalize the challenges were encounter, imagine to measure with Ångstrom resolution the positioning of a protein at a membrane in a sample that contains 10^{11} (< 1 picomol) protein molecules [2]. We achieve this feat, and believe that now is the time to apply this advanced technology to solve veritable biomedical problems.
In the context of this proposal, we demonstrated that the combination of surface-sensitive scattering, *i.e.* NR, with MD simulation provides structural characterization of membrane-associated proteins with atomistic resolution that can be utilized to study the molecular basis of cellular control mechanisms of biological processes, as exemplified in recent work on the PTEN tumor suppressor [3,4]. A major new technical development within the proposed work will be to consistently link experimental scattering results and MD simulation in an integrated modeling approach.

1. D. J. McGillivray, G. Valincius, F. Heinrich, J. W. F. Robertson, D. J. Vanderah, W. Febo-Ayala, I. Ignatjev, M. Lösche, and J. J. Kasianowicz, *Structure of functional Staphylococcus aureus α-hemolysin channels in tethered bilayer lipid membranes*, Biophys. J. 96 (2009), 1547. PMC
2. H. Nanda, S. Datta, F. Heinrich, M. Lösche, A. Rein, S. Krueger, and J. E. Curtis, *Electrostatic interactions and binding orientation of HIV-1 matrix, studied by neutron reflectivity*, Biophys. J. 99 (2010), 2516. PMC
3. S. Shenoy, P. Shekhar, F. Heinrich, M.-C. Daou, A. Gericke, A. H. Ross, and M. Lösche, *Membrane association of the PTEN tumor suppressor: Molecular details of the protein-membrane complex from SPR binding studies and neutron reflection*, PLoS One 7 (2012), e32591. PMC
4. H. Nanda, F. Heinrich, M. Lösche, *Membrane association of the PTEN tumor suppressor: Neutron scattering and MD simulations reveal the structure of protein-membrane complexes*, Methods 77–78 (2015), 136. PMID

B. Positions and Honors

Positions held

1988–1994	Assistant Professor in Physical Chemistry, Chemistry Dept., University of Mainz, Germany.
1994–2005	Associate Professor in Physics, Institute of Exp. Physics I, University of Leipzig, Germany.
1998–2002	Head, Biomembrane Physics group, Institute of Exp. Physics I, University of Leipzig.
2002–2005	Research Professor, Dept. of Chem. Engineering, Johns Hopkins University, Baltimore, MD.
2002–2006	Director, CNBT consortium, NIST Center for Neutron Research (NCNR), Gaithersburg, MD.
2002–present	Associate at the NIST Center for Neutron Research, Gaithersburg, MD.
2005–present	Professor of Physics, Department of Physics, Carnegie Mellon University, Pittsburgh, PA.
2008–present	Professor of Biomedical Engineering (by courtesy), Carnegie Mellon University.

Memberships, Honors, Service

	Fellow, American Physical Society (APS); Member, Biophysical Society (Bethesda, MD), German Biophysical Society (DGfB), American Chemical Society (ACS), Neutron Scattering Society of America (NSSA)
1986–1987	German Research Foundation Postdoctoral Fellow
1988	Guest Lecturer, *École d'été de Physique Théorique*, Les Houches, France
1994	*Habilitation* in Physical Chemistry, Chemistry Department, University of Mainz, Germany
1995	*Universitäts-Professor* (tenure), Institute of Experimental Physics I, University of Leipzig, Germany.
2007	NIH ad-hoc panel review (UO1 Biodefense)
2004–present	Various NSF panel reviews (NIRT program, Bioseparation, BMAT)
2009–2014	Neutron Scattering Science Review Committee, Oak Ridge Natl. Lab., U.S. DOE
2011	Materials Sciences Review Group, Argonne Natl. Lab., Office of Basic Energy Sciences, U.S. DOE
2012	Fellow, American Physical Society
2015	NIH P41 Site Visit Member at Oak Ridge National Laboratory

C. Contributions to Science

(a) One major focus of my research group over many years – and an important anchor point in the layout of this proposal – is the structural characterization of membranes with surface-sensitive scattering techniques. As early as 1990, we pioneered the use of x-ray and neutron reflectometry of floating Langmuir monolayers [1] in which we determined the structural arrangement of phospholipids and the interfacial interaction of proteins with the lipids. Over the years, we developed suitable sample environments and appropriate modeling techniques [2], established rigorous error estimation procedures, improved the sensitivity of the (intrinsically insensitive) neutron measurements and devised strategies to manage beam damage in synchrotron-based x-ray reflectometry. Following my transition to Carnegie Mellon, we moved from focusing on Langmuir monolayers to fully immersed, solid-supported bilayer membranes [3] that show better application potential for addressing biology-related problems. We showed that sparsely-tethered bilayer lipid membranes (stBLMs) can be laterally homogeneous, are exquisitely stable, show extremely low defect density and display fluidity that is comparable with that of lipids in free vesicle membranes [4]. In other words, stBLMs are highly robust model that represent the physical state of the lipid component of biomembranes very well. In addition, they can be characterized with a multitude of physical characterization techniques, including electrochemical impedance spectroscopy (membrane quality) [3], fluorescence correlation spectroscopy (in-plane fluidity) [4] and surface-plasmon spectroscopy (membrane affinity of soluble proteins).

1. D. Vaknin, K. Kjaer, J. Als-Nielsen, and M. Lösche, *Structural properties of DPPC in a monolayer at the air/water interface. A neutron reflection study and reexamination of X-ray reflection experiments*. Biophys. J. 59 (1991), 1325. PMC (130 citations)
2. M. Schalke and M. Lösche, *Structural models of lipid surface monolayers from X-ray and neutron reflectivity measurements*, Adv. Colloid Interf. Sci. 88 (2000), 243. (60 citations)
3. D. J. McGillivray, G. Valincius, D. J. Vanderah, W. Febo-Ayala, J. T. Woodward, F. Heinrich, J. J. Kasianowicz, and M. Lösche, *Molecular-scale structural and functional characterization of sparsely tethered bilayer lipid membranes*, Biointerphases 2 (2007), 21 - 33. (85 citations)
4. S. Shenoy, R. Moldovan, J. Fitzpatrick, D. J. Vanderah, M. Deserno, and M. Lösche, *In-plane homogeneity and lipid dynamics in tethered Bilayer Lipid Membranes (tBLMs*), Soft Matter 6 (2010), 1263. PMC

(b) Following the success in developing model systems that are highly relevant for the investigation of biomembranes, we extended our work to studies of protein-membrane interactions. This work has the potential to close a critical gap in structural biology that is currently not covered by any of the established workhorses – the complexation of intrinsic [1] or peripheral [4] membrane proteins with in-plane fluid, disordered bilayers – because proteins embedded in fluid membranes cannot be crystallized and solid-state NMR techniques are limited to rather small proteins. In distinction, neutron reflection of protein-decorated stBLMs reveals the penetration depth of proteins within a bilayer with Ångstrom precision [1] and determines the orientation of proteins on membranes within a few degrees [4]. In addition, the state of the surface-ligated membrane can be manipulated *in situ*, so that the response of a membrane-ligated protein to external triggers can be characterized (see, for example, Datta, Heinrich, Raghunandan, *et al.*, J. Mol. Biol. 406, 2011, 205). We optimized these systems for sensitivity [3], and demonstrated its utility for a number of biologically relevant problems [1–4]. While the intrinsic resolution of scattering methods is only moderate, combining the experimental results with computational models and, in particular, molecular dynamics simulations is a powerful strategy to extract atomic scale structural information. In addition, the neutron reflection technique holds substantial potential in investigations of multi-protein complexes on membranes in which deuteration of specific protein components enables unraveling the architecture of the complex, as recently shown (Yap, Jiang, Heinrich *et al.*, J. Biol. Chem. 290, 2015, 744).

1. D. J. McGillivray, G. Valincius, F. Heinrich, J. W. F. Robertson, D. J. Vanderah, W. Febo-Ayala, I. Ignatjev, M. Lösche, and J. J. Kasianowicz, *Structure of functional Staphylococcus aureus α-hemolysin channels in tethered bilayer lipid membranes*, Biophys. J. 96 (2009), 1547. PMC (60 citations)
2. G. Valincius, F. Heinrich, R. Budvytyte, D. J. Vanderah, Y. Sokolov, J. E. Hall, and M. Lösche, *Soluble amyloid β oligomers affect dielectric membrane properties by bilayer insertion and domain formation: Implications for cell toxicity*, Biophys. J. 95 (2008), 4845. PMC (105 citations)
3. S. A. Holt, A. P. Le Brun, C. F. Majkrzak, D. J. McGillivray, F. Heinrich, M. Lösche, and J. H. Lakey, *An ion channel containing model membrane: Structural determination by magnetic contrast variation neutron reflectometry*, Soft Matter 5 (2009), 2576. PMC
4. F. Heinrich, H. Nanda, Goh H. Z., C. Bachert, M. Lösche, and A. D. Linstedt, *Myristoylation restricts orientation of the GRASP domain on membranes and promotes membrane tethering,* J. Biol. Chem. 289 (2014), 9683. PMC

(c) As a major player in cell signaling, the PTEN tumor suppressor is frequently mutated in human cancers. PTEN is the antagonist of PI3 kinase in the PI3K/Akt pathway and dephosphorylates the plasma membrane-specific phosphoinositide PI(3,4,5)P_3 at the 3 position of the inositol ring [3]. The mechanistic understanding of PTEN membrane association, substrate turnover and dissociation is still incomplete, but in the past few years, our studies of the PTEN phosphatase on stBLMs and in solution has significantly contributed to filling this gap. Our collaborators Gericke and Ross were able to show that PTEN is allosterically activated by PI(4,5)P_2. We entered the field by working out the details of PTEN association with membranes by combining SPR studies with neutron reflection of the protein on fluid stBLMs [1] and refining the structural model with MD simulations [2]. Recently, we demonstrated with small-angle x-ray scattering (SAXS) that the PTEN phosphatase forms dimers in solution and determined a draft structure of the PTEN homodimer by combining the experimental results with Rosetta docking studies [4].

1. S. Shenoy, P. Shekhar, F. Heinrich, M.-C. Daou, A. Gericke, A. H. Ross, and M. Lösche, *Membrane association of the PTEN tumor suppressor: Molecular details of the protein-membrane complex from SPR binding studies and neutron reflection*, PLoS One 7 (2012), e32591. PMC
2. S. Shenoy, H. Nanda and M. Lösche, *Membrane association of the PTEN tumor suppressor: Electrostatic interaction with phosphatidylserine-containing bilayers and regulatory role of the C-terminal tail*, J. Struct. Biol. 180 (2012), 394. PMC.
3. A. Gericke, N. R. Leslie, M. Lösche, and A. H. Ross, *PtdIns(4,5)P_2-mediated cell Signaling: Emerging principles and PTEN as a paradigm for regulatory mechanism*, Adv. Exp. Med. Biol. 991 (2013), 85. PMC.
4. F. Heinrich, S. Chakravarthy, H. Nanda, A. Papa, P.P. Pandolfi, A.H. Ross, R. K. Harishchandra, A. Gericke, M. Lösche, *The PTEN tumor suppressor forms homodimers in solution*, Structure 23 (2015), 1952 - 1957. PMID

(d) In earlier work, we applied surface-sensitive neutron and x-ray scattering in a variety of problems in materials science, colloidal systems and biotechnology that advanced science and technology in a broad range of areas including polymeric interface films [1,1], liquid-crystalline elastomers [3], and interfacial functionalization with ordered protein arrays [4].

1. J. Schmitt, T. Grünewald, G. Decher, P.S. Pershan, K. Kjaer, and M. Lösche, *Internal structure of layer-by-layer adsorbed polyelectrolyte films: A neutron and x-ray reflectivity study.* Macromolecules 26 (1993) 7058. (350 citations)
2. M. Lösche, J. Schmitt, G. Decher, W.G. Bouwman, and K. Kjaer. *Detailed structure of molecularly thin polyelectrolyte multilayer films on solid substrates as revealed from neutron reflectometry*. Macromolecules 31 (1998), 8893. (440 citations)
3. W. Lehmann, H. Skupin, E. Gebhard, C. Tolksdorf, R. Zentel, P. Krüger, M. Lösche, and F. Kremer, *Giant lateral electrostriction in ferroelectric liquid crystalline elastomers*, Nature 410 (2001), 447. (250 citations)
4. M. Weygand, B. Wetzer, D. Pum, U.B. Sleytr, K. Kjaer, P.B. Howes, and M. Lösche, *Bacterial Surface-layer protein coupling to lipid membranes: X-ray reflectivity and grazing incidence diffraction studies*. Biophys. J. 76 (1999), 458. PMC (85 citations)

(e) A first major contribution to science originated from my PhD work in Germany in the group of H. Möhwald, where I discovered phase separation in floating phospholipid Langmuir monolayers [1] after constructing a dedicated optical microscopy capable of observing label distributions in monomolecular layers on aqueous surfaces [2]. On the one hand, this led to the development of microscopic theories of the long-range interactions in such Langmuir films [3,4] and, on the other hand, preceded the discovery of phase separation in bilayer membranes of giant unilamellar vesicles by two decades (see, for example, Baumgart, Hess & Webb, Nature 425, 2003, 821).

1. M. Lösche, E. Sackmann, and H. Möhwald, *A fluorescence microscopic study concerning the phase diagram of phospholipids*. Ber. Bunsenges. Phys. Chem. 87 (1983), 848. (360 citations)
2. M. Lösche and H. Möhwald, Fluorescence microscope to observe dynamical processes in monomolecular layers at the air/water interface. Rev. Sci. Instrum. 55 (1984), 1968. (210 citations)
3. C.A. Helm, L.A. Laxhuber, M. Lösche, and H. Möhwald, *Electrostatic interactions in phospholipid membranes I: Influence of monovalent ions.* Colloid Polym. Sci. 264 (1986), 46 (150 citations)
4. P. Krüger and M. Lösche, *Molecular chirality and domain shapes in lipid monolayers on aqueous surfaces*, Phys. Rev. E 62 (2000), 7031. (50 citations)

Complete List of Published Work

D. Research Support

Active

2013–2018 NIST-MSE 70NANB13H009, *Innovative Neutron Metrology for the Study of Biomembranes in Health and Disease*, PI: Lösche, US$ 1,146,826.

Goals: Study biomembranes in their physiologically relevant, locally disordered state and develop metrology for their characterization with neutron scattering on the Ångstrom lengthscale.

2011–2016 NIST-MSE 70NANB11H139, *Structure and assembly of membrane-associated proteins*, PI: Lösche, US$ 650,000.

Goals: Develop neutron reflection metrology at the NIST Center for Neutron Research for the investigation of protein-membrane interactions.

2012–2016 NIH 1R01 GM101647-01, *Membrane coupling and dynamic reorganization of Gag in viral budding*, PI: Lösche, US$ 1,163,180.

Goals: Determine the factors that control the association of the Gag MA domain with membranes in binding studies and reveal the mechanisms that lead to the recruitment of specific lipids into the viral shell. Characterize the origin and the implications of the conformational reorganization of full-length Gag at membrane surfaces in experimental and computational studies.

Pending

2016–2020 NIH 1R01 GM101647-01A1, *PTEN Signaling: Membrane Association as a Function of Protein Status*, PI: Lösche, US$ 1,500,966.

Goals: Develop models of the mode of action of the PTEN tumor suppressor as an antagonist of PI3K in the PI3K/Akt pathway. This research will advance our understanding of cancer development and is likely to promote the development of new therapies based upon PTEN-L, a recently discovered secreted form of PTEN.

Completed (past five years)

2008–2013 NIST-MSE 70NANB8H8009, *Ultra-high resolution neutron reflection metrology for the analysis of protein-membrane interactions*, PI: Lösche, US$ 856,460.

Goals: Develop computational tools for the structural assessment of intrinsically disordered proteins and the evaluation of neutron scattering data of such proteins in association with membranes.

2009–2011 NIH 1P01 AG032131, *Membrane-mediated toxicity of* β*-amyloid oligomers*, PI: Lösche, US$ 1,908,742.

Goals: Elucidating the molecular-scale origin of amyloid-related neurodegenerative disease, specifically Alzheimer's. An integrated program that intertwines experimental studies of Aβ oligomer impact on both biomimetic models and cell membranes with molecular-scale computer simulation and membrane theory is designed to shed new light on molecular aspects of cell toxicity of Aβ oligomers.