JCB Journal of Cell Biology

IN MEMORIAM

In Memoriam: Mike Sheetz

Haguy Wolfenson[1], Gregory Giannone[2], and Martin A. Schwartz[3]

Michael P. Sheetz made immense contributions to our understanding of motor proteins, membrane biology, cytoskeleton and mechanobiology over his ~50-year career. He started his independent career at the University of Connecticut, after which he moved to Washington University in St. Louis and then to Duke University, where he led the Department of Cell Biology for 10 years. He then moved to Columbia University, where he established a research group focused on mechanobiology, and then founded and led the Mechanobiology Institute at the National University of Singapore for 10 years. He ended his career at the University of Texas Health Center in Galveston, TX, USA. He trained a generation of leading interdisciplinary cell and mechanobiologists whose independent contributions continue to enhance his legacy.

Michael Patrick Sheetz, one of the preeminent biophysicist/cell biologists of our generation, passed away on January 30 in Galveston, TX, after a long battle with multiple myeloma. Mike was born in Hershey, PA, in 1946 and grew up mainly in Nebraska. He double-majored in chemistry and mathematics at Albion College in Michigan, then did a PhD at Cal Tech with Sunney Chan, where he used nuclear magnetic resonance to study the structure of model lipid bilayers and red blood cell membranes. Mike did postdoctoral research in S.J. Singer's Lab at the University of California, San Diego, then a major center for studies of cell plasma membranes. His work there focused on the red blood cell plasma membrane and cortical cytoskeleton. A productive postdoc led to a faculty position in the Physiology Department at the University of Connecticut School of Medicine in 1974, where he continued work on red blood cell membrane–cytoskeleton interactions, cytoskeletal organization, and small molecular interactions. A growing interest in nonmuscle myosins led to a collaboration with Jim Spudich at Stanford, where they developed an in vitro transport assay to measure myosin motor activity using myosin-coated beads moving on immobilized actin filaments. This assay enabled a collaboration begun at the Marine Biological Labs at Woods Hole with Bruce Schnapp, Ron Vale, and Tom Reese to extend this approach to microtubule motors. Bob Allen from Dartmouth had established microtubule transport assays using the cytoplasm extruded from squid axons. Allen taught the team these assays, which Sheetz et al. used to identify and characterize kinesin-1 as the plus end–directed microtubule motor, distinct from minus end–directed dyneins (Vale et al., 1985). This discovery, so central to modern cell biology and medicine, earned Mike a number of awards, most prominently the Lasker Award, which he shared with Vale and Spudich for their work on motor proteins.

In 1985, he moved to the Department of Cell Biology and Physiology at Washington University in St. Louis as a full professor, then in 1990 to Duke University in North Carolina where he served as Chair of the Department of Cell Biology until 2000, when he moved to Columbia University in New York. While continuing to make major contributions to the microtubule motor field (Kuo and Sheetz, 1993), the Sheetz Lab expanded its interests to study plasma membrane flow and cell adhesion and migration. They published a series of papers that overturned the hypothesis that membrane flow drives cell migration (Kucik et al., 1990) and the first report that lateral membrane protein diffusion is impeded by cytoskeletal barriers (Edidin et al., 1991). A key component of these studies was the adaptation of high-speed cameras to visualize the movement of beads on cell surfaces and laser traps to apply forces to such beads. Sheetz et al. exploited these tools to make a series of seminal discoveries focusing on the behavior of proteins under force. They discovered force-dependent strengthening of integrins (Choquet et al., 1997) and elucidated some of the key mechanisms behind mechanotransduction (Sawada et al., 2006). In a further application, based on the hypothesis derived from structural studies (Papagrigoriou et al., 2004), they showed that force on talin enabled vinculin binding (del Rio et al., 2009). This opened the way to explorations of how cells mechanically probe their environments, from bacteria using type IV pilus machinery to generate forces for twitching motility (Merz et al., 2000), to migrating cells using periodic contractions of their actin-based protrusions (Giannone et al., 2004) or sarcomere-like contractions (Wolfenson et al., 2016) for rigidity sensing. They further studied cytoskeleton–plasma membrane interactions (Raucher et al., 2000) and identified a role for membrane tension in regulation of endocytosis and exocytosis (Raucher

[1]Department of Genetics and Developmental Biology, Rappaport Faculty of Medicine, Technion - Israel Institute of Technology, Haifa, Israel; [2]Interdisciplinary Institute for Neuroscience, Université Bordeaux, CNRS, UMR 5297, Bordeaux, France; [3]Department of Internal Medicine, Department of Cell Biology and Department of Biomedical Engineering, Yale Cardiovascular Research Center, Cardiovascular Medicine, Yale University, New Haven, CT, USA.

Correspondence to Martin A. Schwartz: martin.schwartz@yale.edu.

Mike Sheetz.

and Sheetz, 1999), thus demonstrating homeostatic control of membrane tension. These findings are central to wide areas of cell biology (Iskratsch et al., 2014) and remain active areas of research.

In 2009, Mike moved to Singapore as a founding director of the Mechanobiology Institute (MBI). There, he not only built a great research center but also entered a phase of extraordinary productivity. While maintaining two research groups—at MBI and Columbia University—Mike published a series of remarkable papers, most prominently about integrins and their cytoskeletal connections. These studies elucidated the roles of $\alpha_v\beta_3$ versus $\alpha_5\beta_1$ integrins in mechanotransduction and in supporting matrix forces (Roca-Cusachs et al., 2009), visualized cycles of talin stretch and relaxation in live cells (Margadant et al., 2011), characterized molecular and biophysical mechanisms that govern adhesion (Zhang et al., 2008) and mechanosensing (Changede et al., 2015), and identified differences in mechanosensing between normal and cancer cells (Yang et al., 2020), thereby defining mechanisms and clinical implications for metastasis. In 2019, Mike moved to the University of Texas Medical School in Galveston, continuing research into force, integrins, and cytoskeleton until his death in January.

While the impact of these discoveries was immense, Mike's lab trained leading young scientists whose pioneering work continues his legacy of interdisciplinary innovation. At the Institut Curie in Paris, researchers inspired by his pioneering experiments with optical tweezers used quantitative data from his laboratory to build physical models of how cells generate and respond to mechanical forces. At the Interdisciplinary Institute for Neuroscience in Bordeaux, his early single-molecule tracking experiments shaped new approaches for studying molecular dynamics, not only during cell mechanosensing, but also to track synaptic proteins in neurons. At the Institute for Bioengineering of Catalonia, synergistic experimental modeling studies have further advanced our understanding of cellular mechanosensing. In the IFOM in Milan, scientists have joined forces to understand deregulated mechanosensation in cancer. In the USA, the UK, Japan, China, Singapore, and Israel, his former collaborators, colleagues, and trainees continue to expand studies on adhesion and mechanosensing from basic cell biology, through development and disease.

Mike's lab offered trainees a window into science, wide open to curiosity and discovery. In this space, you had the freedom to exploit state-of-the-art experimental setups and molecular tools to study fundamental biological questions in a quantitative way. As a mentor, he encouraged independence while remaining available as a guide, always ready to steer explorations toward significant breakthroughs. He was a scientist of big and out-of-the-box ideas, which, for members entering the laboratory, could at first be hard to grasp. Nevertheless, Mike fostered a nurturing learning environment that motivated exploration of big ideas, all the while engaging in one-on-one discussions and responding to questions and inquiries with support and encouragement. An important part of the unique environment was the recruitment of people from different disciplines and backgrounds, leading to a range of expertise, viewpoints, and concepts that created a place where people could learn new "languages" and ways of thinking.

His presence in the laboratory was constant and committed. After spending his mornings writing, he would invariably walk through the laboratory, checking on progress with his characteristic "How is it going?" or stopping by during experiments, leaving with an encouraging "I keep my fingers crossed." Encouragement included regular exhortations to productivity with the incantation, "Paper-paper-paper." All the while, Mike maintained a relaxed and easygoing demeanor, with a smile on his face and an approachable presence. Remarkably, during his years at MBI and Columbia University, he somehow seemed to be in both places at once while often traveling across continents. His engagement was truly exceptional. Mike maintained the genuine excitement of a new graduate student when discussing experimental results directly at the microscope or when proposing new experimental strategies.

The most electrifying moments were the whiteboard discussions in his office. Mike's intense, focused gaze could cut through the chatter to detect the faintest signals of discovery. He could see patterns where others only saw noise, linking results into clever conceptual frameworks. His mind worked in multiple dimensions: horizontally, linking ongoing experiments, and vertically, integrating past knowledge with future possibilities. Often, he would bring ideas from disciplines and studies that, at first, seemed unrelated, but turned out to be right on the money. Over the years, he accumulated so much knowledge that it sharpened his mind, embodying Pasteur's famous quote: "In the field of observation, chance only favors the prepared mind." In Mike's lab, you not only witnessed this principle, you lived it.

A striking example occurred at a weekly laboratory meeting when Daniel Choquet presented optical tweezer experiments in which previously trapped beads bound to integrins could no longer be recaptured. While the others laughed at this apparent failure, Mike immediately grasped its significance. He saw evidence of the reinforcement of integrins with the actin cytoskeleton, a key mechanotransduction mechanism. He also recognized that the periodic contraction of actin-based membrane protrusions was a mechanism by which cells sense and

respond to substrate rigidity. His ability to extract new concepts from seemingly unrelated results was truly mind-blowing.

It was remarkable to witness the accumulation of knowledge and expansion of understanding over the years, particularly in the area of integrins and mechanotransduction. The latest development of these ideas was the application of low-frequency ultrasound as a therapeutic tool for diseases such as cancer, osteoarthritis, Alzheimer's disease, and Parkinson's disease. He also found that low-frequency ultrasound could target senescent cells and promote tissue regeneration, showing promise in extending lifespan and activity in mice, and leading to successful phase I clinical trials in osteoarthritis patients, with several other trials currently underway. After his lifetime of achievements, Mike claimed that these studies were the most exciting work of his career.

Mike was driven by movement and action, both in science and in life. An avid tennis player, he shared his passion not only with his wife, Linda Kenney, but also with PhD students and postdocs, forcing them out of the laboratory to practice hitting his backhand or executing precise passing shots. In one of the scientific meetings at MBI, which happened to occur on his 71st birthday, Mike was challenged (or perhaps he challenged himself…) to do 25 pushups in one go! Always energetic and vibrant, he was highly exciting and motivating to be around. Mike is recognized as one of the main architects of molecular mechanobiology. His pioneering work established the fundamental principles of how mechanical forces shape proteins and cells, laying the foundations for an entirely new field. Mike's visionary integration of physics, engineering, and cell biology transformed our understanding of cellular processes, inspiring a generation of scientists and redefining the way we study living systems. We expect that his legacy of discovery, training, and inspiration will live on for at least another generation.

References

Changede, R., X. Xu, F. Margadant, and M.P. Sheetz. 2015. Nascent integrin adhesions form on all matrix rigidities after integrin activation. *Dev. Cell*. 35:614–621. https://doi.org/10.1016/j.devcel.2015.11.001

Choquet, D., D.P. Felsenfeld, and M.P. Sheetz. 1997. Extracellular matrix rigidity causes strengthening of integrin-cytoskeleton linkages. *Cell*. 88: 39–48. https://doi.org/10.1016/S0092-8674(00)81856-5

del Rio, A., R. Perez-Jimenez, R. Liu, P. Roca-Cusachs, J.M. Fernandez, and M.P. Sheetz. 2009. Stretching single talin rod molecules activates vinculin binding. *Science*. 323:638–641. https://doi.org/10.1126/science.1162912

Edidin, M., S.C. Kuo, and M.P. Sheetz. 1991. Lateral movements of membrane glycoproteins restricted by dynamic cytoplasmic barriers. *Science*. 254: 1379–1382. https://doi.org/10.1126/science.1835798

Giannone, G., B.J. Dubin-Thaler, H.G. Döbereiner, N. Kieffer, A.R. Bresnick, and M.P. Sheetz. 2004. Periodic lamellipodial contractions correlate with rearward actin waves. *Cell*. 116:431–443. https://doi.org/10.1016/S0092-8674(04)00058-3

Iskratsch, T., H. Wolfenson, and M.P. Sheetz. 2014. Appreciating force and shape—the rise of mechanotransduction in cell biology. *Nat. Rev. Mol. Cell Biol.* 15:825–833. https://doi.org/10.1038/nrm3903

Kucik, D.F., E.L. Elson, and M.P. Sheetz. 1990. Cell migration does not produce membrane flow. *J. Cell Biol.* 111:1617–1622. https://doi.org/10.1083/jcb.111.4.1617

Kuo, S.C., and M.P. Sheetz. 1993. Force of single kinesin molecules measured with optical tweezers. *Science*. 260:232–234. https://doi.org/10.1126/science.8469975

Margadant, F., L.L. Chew, X. Hu, H. Yu, N. Bate, X. Zhang, and M. Sheetz. 2011. Mechanotransduction in vivo by repeated talin stretch-relaxation events depends upon vinculin. *PLoS Biol.* 9:e1001223. https://doi.org/10.1371/journal.pbio.1001223

Merz, A.J., M. So, and M.P. Sheetz. 2000. Pilus retraction powers bacterial twitching motility. *Nature*. 407:98–102. https://doi.org/10.1038/35024105

Papagrigoriou, E., A.R. Gingras, I.L. Barsukov, N. Bate, I.J. Fillingham, B. Patel, R. Frank, W.H. Ziegler, G.C. Roberts, D.R. Critchley, and J. Emsley. 2004. Activation of a vinculin-binding site in the talin rod involves rearrangement of a five-helix bundle. *EMBO J.* 23:2942–2951. https://doi.org/10.1038/sj.emboj.7600285

Raucher, D., and M.P. Sheetz. 1999. Membrane expansion increases endocytosis rate during mitosis. *J. Cell Biol.* 144:497–506. https://doi.org/10.1083/jcb.144.3.497

Raucher, D., T. Stauffer, W. Chen, K. Shen, S. Guo, J.D. York, M.P. Sheetz, and T. Meyer. 2000. Phosphatidylinositol 4,5-bisphosphate functions as a second messenger that regulates cytoskeleton-plasma membrane adhesion. *Cell*. 100:221–228. https://doi.org/10.1016/S0092-8674(00)81560-3

Roca-Cusachs, P., N.C. Gauthier, A. Del Rio, and M.P. Sheetz. 2009. Clustering of alpha(5)beta(1) integrins determines adhesion strength whereas alpha(v)beta(3) and talin enable mechanotransduction. *Proc. Natl. Acad. Sci. USA*. 106:16245–16250. https://doi.org/10.1073/pnas.0902818106

Sawada, Y., M. Tamada, B.J. Dubin-Thaler, O. Cherniavskaya, R. Sakai, S. Tanaka, and M.P. Sheetz. 2006. Force sensing by mechanical extension of the Src family kinase substrate p130Cas. *Cell*. 127:1015–1026. https://doi.org/10.1016/j.cell.2006.09.044

Vale, R.D., T.S. Reese, and M.P. Sheetz. 1985. Identification of a novel force-generating protein, kinesin, involved in microtubule-based motility. *Cell*. 42:39–50. https://doi.org/10.1016/S0092-8674(85)80099-4

Wolfenson, H., G. Meacci, S. Liu, M.R. Stachowiak, T. Iskratsch, S. Ghassemi, P. Roca-Cusachs, B. O'Shaughnessy, J. Hone, and M.P. Sheetz. 2016. Tropomyosin controls sarcomere-like contractions for rigidity sensing and suppressing growth on soft matrices. *Nat. Cell Biol.* 18:33–42. https://doi.org/10.1038/ncb3277

Yang, B., H. Wolfenson, V.Y. Chung, N. Nakazawa, S. Liu, J. Hu, R.Y. Huang, and M.P. Sheetz. 2020. Stopping transformed cancer cell growth by rigidity sensing. *Nat. Mater.* 19:239–250. https://doi.org/10.1038/s41563-019-0507-0

Zhang, X., G. Jiang, Y. Cai, S.J. Monkley, D.R. Critchley, and M.P. Sheetz. 2008. Talin depletion reveals independence of initial cell spreading from integrin activation and traction. *Nat. Cell Biol.* 10:1062–1068. https://doi.org/10.1038/ncb1765