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On the mobility of biomolecules

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Bibliography

- [1] G. T. Hart, A. K. Ramani, and E. M. Marcotte, “How complete are current yeast and human protein-interaction networks?,” *Genome Biol.*, vol. 7, pp. 120–129, 2006.
- [2] W. Sutherland, “A dynamical theory of diffusion for non-electrolytes and the molecular mass of albumin,” *Phil. Mag.*, vol. 9, pp. 781–785, 1905.
- [3] A. Fick, “Über diffusion,” *Poggendorff’s Annel. Physik*, vol. 94, pp. 59–86, 1855.
- [4] C. Zeiss, *Applications manual LSM 510 ConfoCor 2 fluorescence correlation spectroscopy*. Jena, Germany: Carl Zeiss Advanced Imaging Microscopy, 2001.
- [5] S. Wilhelm, B. Gröbler, M. Gluch, and H. Heinz, *Confocal laser scanning microscopy*. Jena, Germany: Carl Zeiss Advanced Imaging Microscopy, 2003.
- [6] C. Fradin, A. Abu-Arish, R. Granek, and M. Elbaum, “Fluorescence correlation spectroscopy close to a fluctuating membrane,” *Biophys. J.*, vol. 84, pp. 2005–2020, 2003.
- [7] D. Magde, E. L. Elson, and W. W. Webb, “Thermodynamic fluctuations in a reacting system: Measurement by fluorescence correlation spectroscopy,” *Phys. Rev. Lett.*, vol. 29, pp. 705–708, 1972.
- [8] S. R. Aragon and R. Pecora, “Fluorescence correlation spectroscopy as a probe of molecular dynamics,” *J. Chem. Phys.*, vol. 64, pp. 1791–1803, 1976.
- [9] M. K. Doeven, J. H. Folgering, V. Krasnikov, E. R. Geertsma, G. van den Bogaart, and B. Poolman, “Distribution, lateral mobility and function of membrane proteins incorporated into giant unilamellar vesicles,” *Biophys. J.*, vol. 88, pp. 1134–1142, 2005.

- [10] N. Kahya, D. Scherfeld, K. Bacia, B. Poolman, and P. Schwille, "Probing lipid mobility of raft-exhibiting model membranes by fluorescence correlation spectroscopy," *J. Biol. Chem.*, vol. 278, pp. 28109–28115, 2003.
- [11] E. L. Elson and D. Magde, "Fluorescence correlation spectroscopy. I. Conceptual basis and theory," *Biopolymers*, vol. 13, pp. 1–27, 1974.
- [12] P. Schwille, F. J. Meyeralmes, and R. Rigler, "Dual-color fluorescence cross-correlation spectroscopy for multicomponent diffusional analysis in solution," *Biophys. J.*, vol. 72, pp. 1878–1886, 1997.
- [13] G. Veldhuis, M. Hink, V. Krasnikov, G. van den Bogaart, J. Hoeboer, A. J. Visser, J. Broos, and B. Poolman, "The oligomeric state and stability of the mannitol transporter, EnzymeII(mtl), from *Escherichia coli*: a fluorescence correlation spectroscopy study," *Protein Sci.*, vol. 15, pp. 1977–1986, 2006.
- [14] M. K. Doeven, G. van den Bogaart, V. Krasnikov, and B. Poolman, "Probing receptor-translocator interactions in the oligopeptide ABC transporter by fluorescence correlation spectroscopy," *Biophys. J.*, vol. 94, pp. 3956–3965., 2008.
- [15] J. Lippincott-Schwartz, N. Altan-Bonnet, and G. H. Patterson, "Photobleaching and photoactivation: following protein dynamics in living cells," *Nat. Cell Biol.*, vol. 5, pp. S7–S14, 2003.
- [16] J. S. Goodwin and A. K. Kenworthy, "Photobleaching approaches to investigate diffusional mobility and trafficking of Ras in living cells," *Methods*, vol. 37, pp. 154–164, 2005.
- [17] G. van den Bogaart, V. Krasnikov, and B. Poolman, "Dual-color fluorescence-burst analysis to probe protein efflux through the mechanosensitive channel MscL," *Biophys. J.*, vol. 92, pp. 1233–1240, 2007.
- [18] S. I. Sukharev, P. Blount, B. Martinac, F. R. Blattner, and C. Kung, "A large-conductance mechanosensitive channel in *E. coli* encoded by MscL alone," *Nature*, vol. 368, pp. 265–268, 1994.
- [19] G. van den Bogaart, J. T. Mika, V. Krasnikov, and B. Poolman, "The lipid dependence of melittin action investigated by dual-color fluorescence burst analysis," *Biophys. J.*, vol. 93, pp. 154–163, 2007.

- [20] E. Habermann, “Bee and wasp venom: the biochemistry and pharmacology of their peptides and enzymes are reviewed,” *Science*, vol. 177, pp. 314–322, 1972.
- [21] G. van den Bogaart, N. Hermans, V. Krasnikov, A. H. de Vries, and B. Poolman, “On the decrease in lateral mobility of phospholipids by sugars,” *Biophys. J.*, vol. 92, pp. 1598–1605, 2007.
- [22] G. van den Bogaart, N. Hermans, V. Krasnikov, and B. Poolman, “Protein mobility and diffusive barriers in *Escherichia coli*: consequences of osmotic stress,” *Mol. Microbiol.*, vol. 64, pp. 858–871, 2007.
- [23] K. A. Brogden, “Antimicrobial peptides: pore formers or metabolic inhibitors in bacteria?,” *Nat. Rev. Microbiol.*, vol. 3, pp. 238–250, 2005.
- [24] F. Alber, S. Dokudovskaya, L. M. Veenhoff, W. Zhang, J. Kipper, D. Devos, A. Suprpto, O. Karni-Schmidt, R. Williams, B. T. Chait, A. Sali, and M. P. Rout, “The molecular architecture of the nuclear pore complex,” *Nature*, vol. 450, pp. 695–701, 2007.
- [25] R. S. Kass, “The channelopathies: novel insights into molecular and genetic mechanisms of human disease,” *J. Clin. Invest.*, vol. 115, pp. 1986–1989, 2005.
- [26] J. Smisterová, M. van Deemter, G. van der Schaaf, W. Meijberg, and G. Robillard, “Channel protein-containing liposomes as delivery vehicles for the controlled release of drugs - optimization of the lipid composition,” *J. Control. Release*, vol. 101, pp. 382–383, 2005.
- [27] R. E. Hancock and H. G. Sahl, “Antimicrobial and host-defense peptides as new anti-infective therapeutic strategies,” *Nat. Biotechnol.*, vol. 24, pp. 1551–1557, 2006.
- [28] C. C. Cruickshank, R. F. Minchin, A. C. Le Dain, and B. Martinac, “Estimation of the pore size of the large-conductance mechanosensitive ion channel of *Escherichia coli*,” *Biophys. J.*, vol. 73, pp. 1925–1931, 1997.
- [29] L. Movileanu, J. P. Schmittschmitt, J. M. Scholtz, and H. Bayley, “Interactions of peptides with a protein pore,” *Biophys. J.*, vol. 89, pp. 1030–1045, 2005.
- [30] B. A. Krantz, L. Sachs, B. M. Ahlberg, P. Nordin, and T. Nilstun, “A phenylalanine clamp catalyzes protein translocation through the anthrax toxin pore,” *Science*, vol. 309, pp. 777–781, 2005.

- [31] C. Huang, "Studies on phosphatidylcholine vesicles. Formation and physical characteristics," *Biochemistry*, vol. 8, pp. 344–352, 1969.
- [32] M. C. Finkelstein and G. Weissmann, "Enzyme replacement via liposomes. Variations in lipid compositions determine liposomal integrity in biological fluids," *Biochim. Biophys. Acta*, vol. 587, pp. 202–216, 1979.
- [33] T. M. Allen and L. G. Cleland, "Serum-induced leakage of liposome contents," *Biochim. Biophys. Acta Biomembranes*, vol. 597, pp. 418–426, 1980.
- [34] J. N. Weinstein, S. Yoshikami, P. Henkart, R. Blumenthal, and W. A. Hagins, "Liposome-cell interaction: transfer and intracellular release of a trapped fluorescent marker," *Science*, vol. 195, pp. 489–492, 1977.
- [35] Z. Petrásek and P. Schwille, "Precise measurement of diffusion coefficients using scanning fluorescence correlation spectroscopy," *Biophys. J.*, vol. 94, pp. 1437–1448, 2008.
- [36] J. Knol, K. Sjollema, and B. Poolman, "Detergent-mediated reconstitution of membrane proteins," *Biochemistry*, vol. 37, pp. 16410–16415, 1998.
- [37] T. Winkler, J. Ketting, A. Koltermann, and M. Eigen, "Confocal fluorescence coincidence analysis: An approach to ultra high-throughput screening," *Proc. Nat. Acad. Sci. USA*, vol. 96, pp. 1375–1378, 1999.
- [38] K. G. Heinze, M. Rarbach, M. Jahnz, and P. Schwille, "Two-photon fluorescence coincidence analysis: rapid measurements of enzyme kinetics," *Biophys. J.*, vol. 83, pp. 1671–1681, 2002.
- [39] J. Yao, D. R. Larson, H. D. Vishwasrao, W. R. Zipfel, and W. W. Webb, "Blinking and nonradiant dark fraction of water soluble quantum dots in aqueous solution," *Proc. Nat. Acad. Sci. USA*, vol. 102, pp. 14284–14289, 2005.
- [40] H. Li, L. Ying, J. J. Green, S. Balasubramanian, and D. Klenerman, "Ultra-sensitive coincidence fluorescence detection of single DNA molecules," *Anal. Chem.*, vol. 75, pp. 1664–1670, 2003.
- [41] J. J. Green, S. Ladame, L. Ying, D. Klenerman, and S. Balasubramanian, "Investigating a quadruplex-ligand interaction by unfolding kinetics," *J. Am. Chem. Soc.*, vol. 128, pp. 9809–9812, 2006.
- [42] J. R. James, S. S. White, R. W. Clarke, A. M. Johansen, P. D. Dunne, D. L. Sleep, W. J. Fitzgerald, S. J. Davis, and D. Klenerman, "Single-molecule level

- analysis of the subunit composition of the T cell receptor on live T cells,” *Proc. Nat. Acad. Sci. USA*, vol. 104, pp. 17662–17667, 2007.
- [43] K. G. Heinze, A. Koltermann, and P. Schwille, “Simultaneous two-photon excitation of distinct labels for dual-color fluorescence crosscorrelation analysis,” *Proc. Nat. Acad. Sci. USA*, vol. 97, pp. 10377–10382, 2000.
- [44] S. Tristram-Nagle, H. I. Petrache, and J. F. Nagle, “Structure and interactions of fully hydrated dioleoylphosphatidylcholine bi-layers,” *Biophys. J.*, vol. 75, pp. 917–925, 1998.
- [45] N. Kučerka, S. Tristram-Nagle, and J. F. Nagle, “Structure of fully hydrated fluid phase lipid bilayers with monounsaturated chains,” *J. Membrane Biol.*, vol. 208, pp. 1–10, 2005.
- [46] M. A. Agosto, T. Ivanovic, and M. L. Nibert, “Mammalian reovirus, a nonfusogenic nonenveloped virus, forms size-selective pores in a model membrane,” *Proc. Nat. Acad. Sci. USA*, vol. 103, pp. 16496–16501, 2006.
- [47] R. E. Campbell, O. Tour, A. E. Palmer, P. A. Steinbach, G. S. Baird, D. A. Zacharias, and R. Y. Tsien, “A monomeric red fluorescent protein,” *Proc. Nat. Acad. Sci. USA*, vol. 99, pp. 7877–7882, 2002.
- [48] S. J. Shattil, M. Cunningham, T. Wiedmer, J. Zhao, P. J. Sims, and L. F. Brass, “Regulation of glycoprotein IIb-IIIa receptor function studied with platelets permeabilized by the pore-forming complement proteins C5b-9,” *J. Biol. Chem.*, vol. 267, pp. 18424–18431, 1992.
- [49] D. S. Dimitrov and A. E. Sowers, “Membrane electroporation-fast molecular exchange by electroosmosis,” *Biochim. Biophys. Acta*, vol. 1022, pp. 381–392, 1990.
- [50] H. Sauer, L. Pratsch, and R. Peters, “A microassay for the pore-forming activity of complement, perforin, and other cytolytic proteins based on confocal laser scanning microscopy,” *Anal. Biochem.*, vol. 194, pp. 418–424, 1991.
- [51] K. Matsuzaki, S. Yoneyama, and K. Miyajima, “Pore formation and translocation of melittin,” *Biophys. J.*, vol. 73, pp. 831–838, 1997.
- [52] M. P. Bohrer, W. M. Deen, C. R. Robertson, J. L. Troy, and B. M. Brenner, “Influence of molecular configuration on the passage of macromolecules across the glomerular capillary wall,” *J. Gen. Physiol.*, vol. 74, pp. 583–593, 1979.

- [53] E. Gasteiger, C. Hoogland, A. Gattiker, S. Duvaud, M. R. Wilkins, R. D. Appel, and A. Bairoch, *The proteomics protocols handbook*. Heidelberg, Germany: Springer-Verlag, 2005.
- [54] J. P. Colletier, B. Chaize, M. Winterhalter, and D. Fournier, "Protein encapsulation in liposomes: efficiency depends on interactions between protein and phospholipid bilayer," *BMC Biotechnol.*, vol. 2, pp. 9–17, 2002.
- [55] D. Lichtenberg and Y. Barenholz, "Liposomes: preparation, characterization, and preservation," *Methods Biochem. Anal.*, vol. 33, pp. 337–462, 1988.
- [56] C. J. Kirby and G. Gregoriadis, "Preparation of liposomes containing factor VIII for oral treatment of haemophilia," *J. Microencapsul.*, vol. 1, pp. 33–45, 1984.
- [57] A. Moscho, O. Orwar, D. T. Chiu, B. P. Modi, and R. N. Zare, "Rapid preparation of giant unilamellar vesicles," *Proc. Nat. Acad. Sci. USA*, vol. 93, pp. 11443–11447, 1996.
- [58] F. Olson, C. A. Hunt, F. C. Szoka, W. J. Vail, and D. Papahadjopoulos, "Preparation of liposomes of defined size distribution by extrusion through polycarbonate membranes," *Biochim. Biophys. Acta*, vol. 557, pp. 9–23, 1979.
- [59] L. D. Mayer, M. J. Hope, and P. R. Cullis, "Vesicles of variable sizes produced by a rapid extrusion procedure," *Biochim. Biophys. Acta*, vol. 858, pp. 161–168, 1986.
- [60] M. J. Hope, M. B. Bally, G. Webb, and P. R. Cullis, "Production of large unilamellar vesicles by a rapid extrusion procedure: characterization of size, trapped volume and ability to maintain a membrane potential," *Biochim. Biophys. Acta*, vol. 812, pp. 55–65, 1985.
- [61] E. Limpert, W. A. Stahel, and M. Abbt, "Lognormal-distribution; bacterial-populations; sensitivity," *Bioscience*, vol. 51, pp. 341–352, 2001.
- [62] K. G. Heinze, A. Koltermann, and P. Schwille, "Simultaneous two-photon excitation of distinct labels for dual-color fluorescence crosscorrelation analysis," *Proc. Nat. Acad. Sci. USA*, vol. 97, pp. 10377–10382, 2000.
- [63] K. Yoshimura, A. Batiza, and C. Kung, "Chemically charging the pore constriction site opens the mechanosensitive channel MscL," *Biophys. J.*, vol. 80, pp. 2198–2206, 2001.

- [64] M. A. Digman, P. Sengupta, P. W. Wiseman, C. M. Brown, A. R. Horwitz, and E. Gratton, "Fluctuation correlation spectroscopy with a laser-scanning microscope: exploiting the hidden time structure," *Biophys. J.*, vol. 88, pp. 33–36, 2005.
- [65] M. A. Digman, C. M. Brown, P. Sengupta, P. W. Wiseman, A. R. Horwitz, and E. Gratton, "Measuring fast dynamics in solutions and cells with a laser scanning microscope," *Biophys. J.*, vol. 89, pp. 1317–1327, 2005.
- [66] H. Raghuraman and A. Chattopadhyay, "Melittin: a membrane active peptide with diverse functions," *Biosci. Rep.*, vol. 27, pp. 189–223, 2007.
- [67] E. E. Ambroggio, F. Separovic, J. H. Bowie, G. D. Fidelio, and L. A. Bagatolli, "Direct visualization of membrane leakage induced by the antibiotic peptides: maculatin, citropin, and aurein," *Biophys. J.*, vol. 89, pp. 1874–1881, 2005.
- [68] M. T. Lee, W. C. Hung, F. Y. Chen, and H. W. Huang, "Mechanism and kinetics of pore formation in membranes by water-soluble amphipathic peptides," *Proc. Nat. Acad. Sci. USA*, vol. 105, pp. 5087–5092, 2008.
- [69] E. R. Geertsma, N. A. Mahmood, G. K. Schuurman-Wolters, and B. Poolman, "Membrane reconstitution of ABC transporters and assays of translocator function," *Nat. Protoc.*, vol. 3, pp. 256–266, 2008.
- [70] A. J. M. Driessen and N. Nouwen, "Protein translocation across the bacterial cytoplasmic membrane," *Annu. Rev. Biochem.*, vol. 77, pp. 643–667, 2008.
- [71] J. de Keyzer, C. van der Does, T. G. Kloosterman, and A. J. M. Driessen, "Direct demonstration of ATP-dependent release of SecA from a translocating preprotein by surface plasmon resonance," *J. Biol. Chem.*, vol. 278, pp. 29581–29586, 2003.
- [72] N. G. Nossal and L. A. Heppel, "The release of enzymes by osmotic shock from *Escherichia coli* in exponential phase," *J. Biol. Chem.*, vol. 241, pp. 3055–3062, 1966.
- [73] M. Schleyer, R. Schmid, and E. P. Bakker, "Transient, specific and extremely rapid release of osmolytes from growing cells of *Escherichia coli* K-12 exposed to hypoosmotic shock," *Arch. Microbiol.*, vol. 160, pp. 424–431, 1993.
- [74] N. Vázquez-Laslop, H. Lee, R. Hu, and A. A. Neyfakh, "Molecular sieve mechanism of selective release of cytoplasmic proteins by osmotically shocked *Escherichia coli*," *J. Bact.*, vol. 183, pp. 2399–2404, 2001.

- [75] B. Ajouz, C. Berrier, A. Garrigues, M. Besnard, and A. Ghazi, "Release of thioredoxin via mechanosensitive channel MscL during osmotic downshock of *Escherichia coli* cells," *J. Biol. Chem.*, vol. 273, pp. 26670–26674, 1998.
- [76] J. H. A. Folgering, J. M. Kuiper, A. H. de Vries, J. B. F. N. Engberts, and B. Poolman, "Lipid-mediated light activation of a mechanosensitive channel of large conductance," *Langmuir*, vol. 20, pp. 6985–6987, 2004.
- [77] A. Koçer, M. Walko, W. Meijberg, and B. L. Feringa, "A light-actuated nanovalve derived from a channel protein," *Science*, vol. 309, pp. 755–758, 2005.
- [78] S. I. Sukharev, M. Betanzos, C. Chiang, and H. R. Guy, "The gating mechanism of the large mechanosensitive channel MscL," *Nature*, vol. 409, pp. 720–724, 2001.
- [79] J. H. A. Folgering, J. C. Wolters, and B. Poolman, "Engineering covalent oligomers of the mechanosensitive channel of large conductance from *Escherichia coli* with native conductance and gating characteristics," *Protein Sci.*, vol. 14, pp. 2947–2954, 2005.
- [80] S. I. Sukharev, W. J. Sigurdson, C. Kung, and F. Sachs, "Energetic and spatial parameters for gating of the bacterial large conductance mechanosensitive channel, MscL," *J. Gen. Physiol.*, vol. 113, pp. 525–540, 1999.
- [81] F. M. Hantash, M. Ammerlaan, and C. F. Earhart, "Enterobactin synthase polypeptides of *Escherichia coli* are present in an osmotic-shocksensitive cytoplasmic locality," *Microbiology*, vol. 143, pp. 147–156, 1997.
- [82] C. Berrier, A. Garrigues, G. Richarme, and A. Ghazi, "Elongation factor Tu and DnaK are transferred from the cytoplasm to the periplasm of *Escherichia coli* during osmotic downshock presumably via the mechanosensitive channel MscL," *J. Bacteriol.*, vol. 182, pp. 248–251, 2001.
- [83] G. R. Jacobson, B. J. Takacs, and J. P. Rosenbusch, "Properties of a major protein released from *Escherichia coli* by osmotic shock," *Biochemistry*, vol. 15, pp. 2297–2303, 1976.
- [84] P. Blount, S. I. Sukharev, P. C. Moe, M. J. Schröder, H. R. Guy, and C. Kung, "Membrane topology and multimeric structure of a mechanosensitive channel protein of *Escherichia coli*," *EMBO J.*, vol. 15, pp. 4798–4805, 1996.

- [85] X. Ou, P. Blount, R. Hoffman, and C. Kung, "One face of a transmembrane helix is crucial in mechanosensitive gating," *Proc. Nat. Acad. Sci. USA*, vol. 95, pp. 11471–11475, 1998.
- [86] R. Rigler, U. Mets, J. Widengren, and P. Kask, "Fluorescence correlation spectroscopy with high count rate and low background: analysis of translational diffusion," *Eur. Biophys. J.*, vol. 22, pp. 169–175, 1993.
- [87] J. Reizer, S. L. Sutrina, L. F. Wu, J. Deutscher, R. Reddy, and M. H. Saier Jr., "Functional interactions between proteins of the phosphoenolpyruvate : sugar phosphotransferase systems of *Bacillus subtilis* and *Escherichia coli*," *J. Biol. Chem.*, vol. 267, pp. 9158–9169, 1992.
- [88] K. Bacia and P. Schwille, "A dynamic view of cellular processes by *in vivo* fluorescence auto- and cross-correlation spectroscopy," *Methods*, vol. 29, pp. 74–85, 2003.
- [89] P. K. Hammen, E. B. Waygood, and R. E. Kleivit, "Reexamination of the secondary and tertiary structure of histidine-containing protein from *Escherichia coli* by homonuclear and heteronuclear NMR spectroscopy," *Biochemistry*, vol. 30, pp. 11842–11850, 1991.
- [90] N. Carulla, C. Woodward, and G. Barany, "Synthesis and characterization of a beta-hairpin peptide that represents a 'core module' of bovine pancreatic trypsin inhibitor (BPTI)," *Biochemistry*, vol. 39, pp. 7927–7937, 2000.
- [91] H. Reutimann, B. Straub, P. L. Luisi, and A. Holmgren, "A conformational study of thioredoxin and its tryptic fragments," *J. Biol. Chem.*, vol. 256, pp. 6796–6803, 1981.
- [92] R. L. Hill, K. Brew, T. C. Vanaman, I. P. Trayer, and P. Mattock, "The structure, function, and evolution of alpha-lactalbumin," *Brookhaven. Symp. Biol.*, vol. 21, pp. 139–154, 1968.
- [93] P. D. Jeffrey and J. H. Coates, "An equilibrium ultracentrifuge study of the self-association of bovine insulin," *Biochemistry*, vol. 5, pp. 489–498, 1966.
- [94] S. Ludvigsen, H. B. Olsen, and N. C. Kaarsholm, "A structural switch in a mutant insulin exposes key residues for receptor binding," *J. Mol. Biol.*, vol. 279, pp. 1–7, 1998.

- [95] S. Parkin, B. Rupp, and H. Hope, "Structure of bovine pancreatic trypsin inhibitor at 125 K definition of carboxyl-terminal residues Gly57 and Ala58," *Acta Crystallogr. D. Biol. Crystallogr.*, vol. 52, pp. 18–29, 1996.
- [96] S. Napper, L. T. J. Delbaere, and E. B. Waygood, "The aspartyl replacement of the active site histidine-containing protein, HPr, of the *Escherichia coli* phosphoenolpyruvate : sugar phosphotransferase system can accept and donate a phosphoryl group. Spontaneous dephosphorylation of acyl-phosphate autocatalyzes an internal cyclization," *J. Biol. Chem.*, vol. 274, pp. 21776–21782, 1999.
- [97] B. W. Lennon, C. H. Williams Jr., and M. L. Ludwig, "Crystal structure of reduced thioredoxin reductase from *Escherichia coli*: structural flexibility in the isoalloxazine ring of the flavin adenine dinucleotide cofactor," *Protein Sci.*, vol. 8, pp. 2366–2379, 1999.
- [98] N. Chandra, K. Brew, and K. R. Acharya, "Structural evidence for the presence of a secondary calcium binding site in human α -lactalbumin," *Biochemistry*, vol. 37, pp. 4767–4772, 1998.
- [99] A. Anishkin, V. Gendel, N. A. Sharifi, C. S. Chiang, L. Shirinian, H. R. Guy, and S. Sukharev, "On the conformation of the COOH-terminal domain of the large mechanosensitive channel MscL," *J. Gen. Physiol.*, vol. 121, pp. 227–244, 2003.
- [100] J. H. A. Folgering, P. C. Moe, G. K. Schuurman-Wolters, P. Blount, and B. Poolman, "*Lactococcus lactis* uses MscL as its principal mechanosensitive channel," *J. Biol. Chem.*, vol. 280, pp. 8784–8792, 2005.
- [101] P. Blount, S. I. Sukharev, M. J. Schröder, S. K. Nagle, and C. Kung, "Single residue substitutions that change the gating properties of a mechanosensitive channel in *Escherichia coli*," *Proc. Nat. Acad. Sci. USA*, vol. 93, pp. 11652–11657, 1996.
- [102] P. Moe and P. Blount, "Assessment of potential stimuli for mechano-dependent gating of MscL: effects of pressure, tension, and lipid headgroups," *Biochemistry*, vol. 44, pp. 12239–12244, 2005.
- [103] C. C. Hase, A. C. Le Dain, and B. Martinac, "Purification and functional reconstitution of the recombinant large mechanosensitive ion channel (MscL) of *Escherichia coli*," *J. Biol. Chem.*, vol. 270, pp. 18329–18334, 1995.

- [104] A. Koçer, M. Walko, E. Bulten, E. Halza, B. L. Feringa, and W. Meijberg, "Rationally designed chemical modulators convert a bacterial channel protein into a pH-sensory valve," *Angew. Chem. Int. Ed.*, vol. 45, pp. 3126–3130, 2006.
- [105] O. Toke, "Antimicrobial peptides: new candidates in the fight against bacterial infections," *Biopolymers*, vol. 80, pp. 717–735, 2005.
- [106] B. B. Finlay and R. E. Hancock, "Can innate immunity be enhanced to treat microbial infections?," *Nat. Rev. Microbiol.*, vol. 2, pp. 497–504, 2004.
- [107] R. E. Hancock and A. Patrzykat, "Clinical development of cationic antimicrobial peptides: from natural to novel antibiotics," *Curr. Drug Targets Infect. Disord.*, vol. 2, pp. 79–83, 2002.
- [108] A. S. Ladokhin and S. H. White, "'detergent-like' permeabilization of anionic lipid vesicles by melittin," *Biochim. Biophys. Acta*, vol. 1514, pp. 253–260, 2001.
- [109] J. Lauterwein, L. R. Brown, and K. Wuthrich, "High-resolution 1H-NMR studies of monomeric melittin in aqueous solution," *Biochim. Biophys. Acta*, vol. 622, pp. 219–230, 1980.
- [110] J. C. Talbot, J. Dufourcq, J. de Bony, J. F. Faucon, and C. Lussan, "Conformational change and self association of monomeric melittin," *FEBS Lett.*, vol. 102, pp. 191–193, 1979.
- [111] D. Allende, S. A. Simon, and T. J. McIntosh, "Melittin-induced bilayer leakage depends on lipid material properties: evidence for toroidal pores," *Biophys. J.*, vol. 88, pp. 1828–1837, 2005.
- [112] M. Iwadate, T. Asakura, and M. P. Williamson, "The structure of the melittin tetramer at different temperatures an NOE-based calculation with chemical shift refinement," *Eur. J. Biochem.*, vol. 257, pp. 479–487, 1998.
- [113] F. Inagaki, I. Shimada, K. Kawaguchi, M. Hirano, I. Terasawa, T. Ikura, and N. Go, "Structure of melittin bound to perdeuterated dodecylphosphocholine micelles as studied by two-dimensional NMR and distance geometry calculations," *Biochemistry*, vol. 28, pp. 5985–5991, 1989.
- [114] T. Ikura, N. Go, and F. Inagaki, "Refined structure of melittin bound to perdeuterated dodecylphosphocholine micelles as studied by 2D-NMR and distance geometry calculation," *Proteins*, vol. 9, pp. 81–89, 1991.

- [115] H. Vogel and F. Jahnig, "The structure of melittin in membranes.," *Biophys. J.*, vol. 50, pp. 573–582, 1986.
- [116] S. S. Saini, A. K. Chopra, and J. W. Peterson, "Melittin activates endogenous phospholipase D during cytolysis of human monocytic leukemia cells," *Toxicon*, vol. 37, pp. 1605–1619, 1999.
- [117] S. S. Saini, A. K. Chopra, and J. W. Peterson, "Melittin-mediated release of [3H]-oleic acid from *E. coli* cells is dependent upon heat- and trypsin-sensitive factor(s) in human serum," *Toxicon*, vol. 38, pp. 1077–1086, 2000.
- [118] Z. Oren and Y. Shai, "Selective lysis of bacteria but not mammalian cells by diastereomers of melittin: structure-function study," *Biochemistry*, vol. 36, pp. 1826–1835, 1997.
- [119] Y. Shai and Z. Oren, "Diastereoisomers of cytolysins, a novel class of potent antibacterial peptides," *J. Biol. Chem.*, vol. 271, pp. 7305–7308, 1996.
- [120] M. T. Lee, F. Y. Chen, and H. W. Huang, "Energetics of pore formation induced by membrane active peptides," *Biochemistry*, vol. 43, pp. 3590–3599, 2004.
- [121] L. Yang, T. A. Harroun, T. M. Weiss, L. Ding, and H. W. Huang, "Barrel-stave model or toroidal model? A case study on melittin pores," *Biophys. J.*, vol. 81, pp. 1475–1485, 2001.
- [122] M. P. Sansom, "The biophysics of peptide models of ion channels," *Prog. Biophys. Mol. Biol.*, vol. 55, pp. 139–215, 1991.
- [123] A. Naito, T. Nagao, K. Norisada, T. Mizuno, S. Tuzi, and H. Saito, "Conformation and dynamics of melittin bound to magnetically oriented lipid bilayers by solid-state (31)P and (13)C NMR spectroscopy," *Biophys. J.*, vol. 78, pp. 2405–2417, 2000.
- [124] R. Smith, F. Separovic, T. J. Milne, A. Whittaker, F. M. Bennett, B. A. Cornell, and A. Makriyannis, "Structure and orientation of the pore-forming peptide, melittin, in lipid bilayers," *J. Mol. Biol.*, vol. 241, pp. 456–466, 1994.
- [125] Y. H. Lam, S. R. Wassall, C. J. Morton, R. Smith, and F. Separovic, "Solid-state NMR structure determination of melittin in a lipid environment," *Biophys. J.*, vol. 81, pp. 2752–2761, 2001.

- [126] S. J. Ludtke, K. He, W. T. Heller, T. A. Harroun, L. Yang, and H. W. Huang, "Membrane pores induced by magainin," *Biochemistry*, vol. 35, pp. 13723–13728, 1996.
- [127] M. J. Gómara, S. Nir, and J. L. Nieva, "Effects of sphingomyelin on melittin pore formation," *Biochim. Biophys. Acta*, vol. 1612, pp. 83–99, 2003.
- [128] K. Hristova, C. E. Dempsey, and S. H. White, "Structure, location, and lipid perturbations of melittin at the membrane interface," *Biophys. J.*, vol. 80, pp. 801–811, 2001.
- [129] M. J. Citra and P. H. Axelsen, "Determination of molecular order in supported lipid membranes by internal reflection Fourier transform infrared spectroscopy," *Biophys. J.*, vol. 71, pp. 1796–1805, 1996.
- [130] C. Altenbach, W. Froncisz, J. S. Hyde, and W. L. Hubbell, "Conformation of spin-labeled melittin at membrane surfaces investigated by pulse saturation recovery and continuous wave power saturation electron paramagnetic resonance," *Biophys. J.*, vol. 56, pp. 1183–1191, 1989.
- [131] B. Stanislawski and H. Rüterjans, "¹³C-NMR investigation of the insertion of the bee venom melittin into lecithin vesicles," *Eur. Biophys. J.*, vol. 15, pp. 1–15, 1987.
- [132] C. E. Dempsey and G. S. Butler, "Helical structure and orientation of melittin in dispersed phospholipid membranes from amide exchange analysis *in situ*," *Biochemistry*, vol. 31, pp. 11973–11977, 1992.
- [133] J. Dufourcq, J. F. Faucon, G. Fourche, J. L. Dasseux, M. le Maire, and T. Gulik-Krzywicki, "Morphological changes of phosphatidylcholine bilayers induced by melittin: vesicularization, fusion, discoidal particles," *Biochim. Biophys. Acta*, vol. 859, pp. 33–48, 1986.
- [134] J. H. Kleinschmidt, J. E. Mahaney, D. D. Thomas, and D. Marsh, "Interaction of bee venom melittin with zwitterionic and negatively charged phospholipid bilayers: a spin-label electron spin resonance study," *Biophys. J.*, vol. 72, pp. 767–778, 1997.
- [135] A. S. Ladokhin, M. E. Selsted, and S. H. White, "Sizing membrane pores in lipid vesicles by leakage of co-encapsulated markers: pore formation by melittin," *Biophys. J.*, vol. 72, pp. 1762–1766, 1997.

- [136] S. C. Park, J. Y. Kim, S. O. Shin, C. Y. Jeong, M. H. Kim, S. Y. Shin, G. W. Cheong, Y. Park, and K. S. Hahm, "Investigation of toroidal pore and oligomerization by melittin using transmission electron microscopy," *Biochem. Biophys. Res. Commun.*, vol. 343, pp. 222–228, 2006.
- [137] K. Matsuzaki, O. Murase, and K. Miyajima, "Kinetics of pore formation by an antimicrobial peptide, magainin 2, in phospholipid bilayers," *Biochemistry*, vol. 34, pp. 12553–12559, 1995.
- [138] K. Hristova, M. E. Selsted, and S. H. White, "Critical role of lipid composition in membrane permeabilization by rabbit neutrophil defensins," *J. Biol. Chem.*, vol. 272, pp. 24224–24233, 1997.
- [139] J. H. Kang, S. Y. Shin, S. Y. Jang, M. K. Lee, and K. S. Hahm, "Release of aqueous contents from phospholipid vesicles induced by cecropin A (1-8)-magainin 2 (1-12) hybrid and its analogues," *J. Pept. Res.*, vol. 52, pp. 45–50, 1998.
- [140] H. Zhao, J. P. Mattila, J. M. Holopainen, and P. K. Kinnunen, "Comparison of the membrane association of two antimicrobial peptides, magainin 2 and indolicidin," *Biophys. J.*, vol. 81, pp. 2979–2991, 2001.
- [141] T. Benachir and M. Lafleur, "Study of vesicle leakage induced by melittin," *Biochim. Biophys. Acta*, vol. 1235, pp. 452–460, 1995.
- [142] T. Benachir and M. Lafleur, "Osmotic and pH transmembrane gradients control the lytic power of melittin," *Biophys. J.*, vol. 70, pp. 831–840, 1996.
- [143] T. Katsu, C. Ninomiya, M. Kuroko, H. Kobayashi, T. Hirota, and Y. Fujita, "Action mechanism of amphipathic peptides gramicidin S and melittin on erythrocyte membrane," *Biochim. Biophys. Acta*, vol. 939, pp. 57–63, 1988.
- [144] T. C. Laurent and K. A. Granath, "Fractionation of dextran and ficoll by chromatography on sephadex G-200," *Biochim. Biophys. Acta*, vol. 136, pp. 191–198, 1967.
- [145] S. W. Hui, T. P. Stewart, P. L. Yeagle, and A. D. Albert, "Bilayer to non-bilayer transition in mixtures of phosphatidylethanolamine and phosphatidylcholine: implications for membrane properties," *Arch. Biochem. Biophys.*, vol. 207, pp. 227–240, 1981.
- [146] D. S. Goodsell, "Inside a living cell," *Trends Biochem. Sci.*, vol. 16, pp. 203–206, 1991.

- [147] E. Biemans-Oldehinkel and B. Poolman, "On the role of the two extracytoplasmic substrate-binding domains in the ABC transporter OpuA," *EMBO J.*, vol. 22, pp. 5983–5993, 2003.
- [148] B. R. Terry, E. K. Matthews, and J. Haseloff, "Molecular characterisation of recombinant green fluorescent protein by fluorescence correlation microscopy," *Biochem. Biophys. Res. Commun.*, vol. 217, pp. 21–27, 1995.
- [149] R. Smith, F. Separovic, F. C. Bennett, and B. A. Cornell, "Melittin-induced changes in lipid multilayers. A solid-state NMR study," *Biophys. J.*, vol. 63, pp. 469–474, 1992.
- [150] A. M. Batenburg, J. H. van Esch, and B. de Kruijff, "Melittin-induced changes of the macroscopic structure of phosphatidylethanolamines," *Biochemistry*, vol. 27, pp. 2324–2331, 1988.
- [151] C. G. Morgan, H. Williamson, S. Fuller, and B. Hudson, "Melittin induces fusion of unilamellar phospholipid vesicles," *Biochim. Biophys. Acta*, vol. 732, pp. 668–674, 1983.
- [152] Y. Higashino, A. Matsui, and K. Ohki, "Membrane fusion between liposomes composed of acidic phospholipids and neutral phospholipids induced by melittin: a differential scanning calorimetric study," *J. Biochem. (Tokyo)*, vol. 130, pp. 393–397, 2001.
- [153] G. D. Eytan and T. Almary, "Melittin-induced fusion of acidic liposomes," *FEBS Lett.*, vol. 156, pp. 29–32, 1983.
- [154] M. Monette, M. R. van Calsteren, and M. Lafleur, "Effect of cholesterol on the polymorphism of dipalmitoylphosphatidylcholine / melittin complexes: an NMR study," *Biochim. Biophys. Acta*, vol. 1149, pp. 319–328, 1993.
- [155] N. Papo and Y. Shai, "Exploring peptide membrane interaction using surface plasmon resonance: differentiation between pore formation versus membrane disruption by lytic peptides," *Biochemistry*, vol. 42, pp. 458–466, 2003.
- [156] T. Benachir, M. Monette, J. Grenier, and M. Lafleur, "Melittin-induced leakage from phosphatidylcholine vesicles is modulated by cholesterol: a property used for membrane targeting," *Eur. Biophys. J.*, vol. 25, pp. 201–210, 1997.
- [157] H. Raghuraman and A. Chattopadhyay, "Interaction of melittin with membrane cholesterol: a fluorescence approach," *Biophys. J.*, vol. 87, pp. 2419–2432, 2004.

- [158] D. K. Hinch and J. H. Crowe, "The lytic activity of the bee venom peptide melittin is strongly reduced by the presence of negatively charged phospholipids or chloroplast galactolipids in the membranes of phosphatidylcholine large unilamellar vesicles," *Biochim. Biophys. Acta*, vol. 1284, pp. 162–170, 1996.
- [159] H. W. Huang, "Molecular mechanism of antimicrobial peptides: The origin of cooperativity," *Biochim. Biophys. Acta*, vol. 1758, pp. 1292–1302, 2006.
- [160] K. Hristova, W. C. Wimley, V. K. Mishra, G. P. Anantharamiah, J. P. Segrest, and S. H. White, "An amphipathic alpha-helix at a membrane interface: a structural study using a novel X-ray diffraction method," *J. Mol. Biol.*, vol. 290, pp. 99–117, 1999.
- [161] Y. Wu, K. He, S. J. Ludtke, and H. W. Huang, "X-ray diffraction study of lipid bilayer membranes interacting with amphiphilic helical peptides: diphytanoyl phosphatidylcholine with alamethicin at low concentrations," *Biophys. J.*, vol. 68, pp. 2361–2369, 1995.
- [162] R. E. Jacobs and S. H. White, "The nature of the hydrophobic binding of small peptides at the bilayer interface: implications for the insertion of transbilayer helices," *Biochemistry*, vol. 28, pp. 3421–3437, 1989.
- [163] J. Dufourcq, B. Clin, and B. Lemanceau, "NMR study of ganglion-blocking and curare-like dimethoniums conformation in aqueous solutions," *FEBS Lett.*, vol. 22, pp. 205–209, 1972.
- [164] E. J. Dufourc, I. C. Smith, and J. Dufourcq, "Molecular details of melittin-induced lysis of phospholipid membranes as revealed by deuterium and phosphorus NMR," *Biochemistry*, vol. 25, pp. 6448–6455, 1986.
- [165] M. Monette and M. Laffeur, "Influence of lipid chain unsaturation on melittin-induced micellization," *Biophys. J.*, vol. 70, pp. 2195–2202, 1996.
- [166] T. Pott, M. Paternostre, and E. J. Dufourc, "A comparative study of the action of melittin on sphingomyelin and phosphatidylcholine bilayers," *Eur. Biophys. J.*, vol. 27, pp. 237–245, 1998.
- [167] R. Carmieli, N. Papo, H. Zimmermann, A. Potapov, Y. Shai, and D. Goldfarb, "Utilizing ESEEM spectroscopy to locate the position of specific regions of membrane-active peptides within model membranes," *Biophys. J.*, vol. 90, pp. 492–505, 2006.

- [168] K. Hristova, M. E. Selsted, and S. H. White, "Interactions of monomeric rabbit neutrophil defensins with bilayers: comparison with dimeric human defensin HNP-2," *Biochemistry*, vol. 35, pp. 11888–11894, 1996.
- [169] J. H. Crowe, J. F. Carpenter, and L. M. Crowe, "The role of vitrification in anhydrobiosis," *Annu. Rev. Physiol.*, vol. 60, pp. 73–103, 1997.
- [170] T. Hottiger, P. Schmutz, and A. Wiemken, "Heat-induced accumulation and futile cycling of trehalose in *Saccharomyces cerevisiae*," *J. Bacteriol.*, vol. 169, pp. 5518–5522, 1987.
- [171] C. G. Hounsa, E. V. Brandt, J. Thevelein, S. Hohmann, and B. A. Prior, "Role of trehalose in survival of *Saccharomyces cerevisiae* under osmotic stress," *Microbiology*, vol. 144, pp. 671–680, 1998.
- [172] W. Wonisch, M. Hayn, R. J. Schaur, F. Tatzber, I. Kranner, D. Grill, R. Winkler, T. Bilinski, S. D. Kohlwein, and H. Esterbauer, "Increased stress parameter synthesis in the yeast *Saccharomyces cerevisiae* after treatment with 4-hydroxy-2-nonenal," *FEBS Lett.*, vol. 405, pp. 11–15, 1997.
- [173] A. E. Oliver, D. K. Hinch, and J. H. Crowe, "Looking beyond sugars: the role of amphiphilic solutes in preventing adventitious reactions in anhydrobiotes at low water contents," *Comp. Biochem. Physiol. A. Mol. Integr. Physiol.*, vol. 131, pp. 515–525, 2002.
- [174] J. H. Crowe, F. A. Hoekstra, and L. M. Crowe, "Anhydrobiosis," *Annu. Rev. Physiol.*, vol. 54, pp. 579–599, 1992.
- [175] F. A. Hoekstra, E. A. Golovina, F. A. Tetteroo, and W. F. Wolkers, "Induction of desiccation tolerance in plant somatic embryos: how exclusive is the protective role of sugars?," *Cryobiology*, vol. 43, pp. 140–150, 2001.
- [176] D. L. Wright, A. Eroglu, M. Toner, and T. L. Toth, "Use of sugars in cryopreserving human oocytes," *Reprod. Biomed. Online.*, vol. 9, pp. 179–186, 2004.
- [177] J. H. Crowe, L. M. Crowe, A. E. Oliver, N. Tsvetkova, W. Wolkers, and F. Tablin, "The trehalose myth revisited: introduction to a symposium on stabilization of cells in the dry state," *Cryobiology*, vol. 43, pp. 89–105, 2001.
- [178] K. L. Koster, Y. P. Lei, M. Anderson, S. Martin, and G. Bryant, "Effects of vitrified sugars on phosphatidyl choline fluid-to-gel phase transitions," *Biophys. J.*, vol. 78, pp. 1932–1948, 2000.

- [179] C. Cacela and D. K. Hinch, “Low amounts of sucrose are sufficient to depress the phase transition temperature of dry phosphatidylcholine, but not for lyoprotection of liposomes,” *Biophys. J.*, vol. 90, pp. 2831–2842, 2006.
- [180] K. Roe and T. P. Labuza, “Transition and crystallization of amorphous trehalose-sucrose mixtures,” *J. Food Properties*, vol. 9, pp. 1–18, 2006.
- [181] J. H. Crowe, L. M. Crowe, J. F. Carpenter, A. S. Rudolph, A. C. Wistrom, B. J. Spargo, and T. J. Anchordoguy, “Interactions of sugars with membranes,” *Biochim. Biophys. Acta*, vol. 947, pp. 367–384, 1988.
- [182] L. M. Crowe, R. Mouradian, J. H. Crowe, S. A. Jackson, and C. Womersley, “Effects of carbohydrates on membrane stability at low water activities,” *Biochim. Biophys. Acta*, vol. 769, pp. 141–150, 1984.
- [183] D. K. Hinch, E. Zuther, E. M. Hellwege, and A. G. Heyer, “Specific effects of fructo- and gluco-oligosaccharides in the preservation of liposomes during drying,” *Glycobiology*, vol. 12, pp. 103–110, 2002.
- [184] M. Luzardo, F. Amalfa, A. M. Nunez, A. C. Biondi de Lopez, and E. A. Disalvo, “Effect of trehalose and sucrose on the hydration and dipole potential of lipid bilayers,” *Biophys. J.*, vol. 78, pp. 2452–2458, 2000.
- [185] L. M. Crowe and J. H. Crowe, “Solution effects on the thermotropic phase transition of unilamellar liposomes,” *Biochim. Biophys. Acta*, vol. 1064, pp. 267–274, 1991.
- [186] S. Ohtake, C. Schebor, and J. J. de Pablo, “Effects of trehalose on the phase behavior of DPPC-cholesterol unilamellar vesicles,” *Biochim. Biophys. Acta*, vol. 1758, pp. 65–73, 2006.
- [187] D. van der Spoel, E. Lindahl, B. Hess, A. R. van Buuren, E. Apol, P. J. Meulenhoff, D. P. Tieleman, A. L. T. M. Sijbers, K. A. Feenstra, R. van Drunen, and H. J. C. Berendsen, *GROMACS user manual version 3.2*. <http://www.gromacs.org>, 2004.
- [188] C. Oostenbrink, A. Villa, A. E. Mark, and W. F. van Gunsteren, “A biomolecular force field based on the free enthalpy of hydration and solvation: the GROMOS force-field parameter sets 53A5 and 53A6,” *J. Comput. Chem.*, vol. 25, pp. 1656–1676, 2004.

- [189] H. J. C. Berendsen, J. P. M. Postma, W. F. van Gunsteren, and J. Hermans, *Interaction models for water in relation to protein hydration*. Dordrecht, The Netherlands: Reidel Publishing Company, 1981.
- [190] R. W. Hockney, "The potential calculation and some applications," *Meth. Comput. Phys.*, vol. 9, pp. 136–211, 1970.
- [191] B. Hess, H. Bekker, H. J. C. Berendsen, and J. G. E. M. Fraaije, "LINCS: a linear constraint solver for molecular simulations," *J. Comput. Chem.*, vol. 18, pp. 1463–1472, 1997.
- [192] S. Miyamoto and P. A. Kollman, "SETTLE: an analytical version of the SHAKE and RATTLE algorithm for rigid water models," *J. Comput. Chem.*, vol. 13, pp. 952–962, 1992.
- [193] H. J. C. Berendsen, J. P. M. Postma, A. DiNola, and J. R. Haak, "Molecular dynamics with coupling to an external bath," *J. Chem. Phys.*, vol. 81, pp. 3684–3690, 1984.
- [194] I. G. Tironi, R. Sperb, P. E. Smith, and W. F. van Gunsteren, "A generalized reaction field method for molecular dynamics simulations," *J. Chem. Phys.*, vol. 102, pp. 5451–5459, 1995.
- [195] A. H. de Vries, A. E. Mark, and S. J. Marrink, "The binary mixing behavior of phospholipids in a bi-layer: a molecular dynamics study," *J. Phys. Chem. B.*, vol. 108, pp. 2454–2463, 2004.
- [196] C. S. Pereira, R. D. Lins, I. Chandrasekhar, L. C. G. Freitas, and P. H. Hünenberger, "Interaction of the disaccharide trehalose with a phospholipid bi-layer: a molecular dynamics study," *Biophys. J.*, vol. 86, pp. 2273–2285, 2004.
- [197] H. M. Berman, J. Westbrook, Z. Feng, G. Gilliland, T. N. Bhat, H. Weissig, I. N. Shindyalov, and P. E. Bourne, "The protein data bank," *Nucleic Acids Res.*, vol. 28, pp. 235–242, 2000.
- [198] A. W. Schüttelkopf and D. M. F. van Aalten, "PRODRG - a tool for high-throughput crystallography of protein-ligand complexes," *Acta Crystallogr. D Biol. Crystallogr.*, vol. 60, pp. 1355–1363, 2004.
- [199] R. D. Lins and P. H. Hünenberger, "A new GROMOS parameter set for hexopyranose-based carbohydrates," *J. Comput. Chem.*, vol. 26, pp. 1400–1412, 2005.

- [200] S. J. Marrink, A. H. de Vries, and A. E. Mark, "Coarse grained model for semiquantitative lipid simulations," *J. Phys. Chem. B.*, vol. 108, pp. 750–760, 2004.
- [201] A. Fillipov, G. Oradd, and G. Lindblom, "The effect on cholesterol on the lateral diffusion of phospholipids in oriented bilayers," *Biophys. J.*, vol. 84, pp. 3079–3086, 2003.
- [202] P. G. Saffman and M. Delbrück, "Brownian motion in biological membranes," *Proc. Nat. Acad. Sci. USA*, vol. 72, pp. 3111–3113, 1975.
- [203] A. Skibinsky, R. M. Venable, and R. W. Pastor, "A molecular dynamics study of the response of lipid bi-layers and monolayers to trehalose," *Biophys. J.*, vol. 89, pp. 4111–4121, 2005.
- [204] A. K. Sum, R. Faller, and J. J. de Pablo, "Molecular simulation study of phospholipid bi-layers and insights of the interactions with disaccharides," *Biophys. J.*, vol. 85, pp. 2830–2844, 2003.
- [205] M. C. Wiener and S. H. White, "Structure of a fluid dioleoylphosphatidylcholine bi-layer determined by joint refinement of X-ray and neutron diffraction data. II. Distribution and packing of terminal methyl groups," *Biophys. J.*, vol. 61, pp. 428–433, 1992.
- [206] I. Chandrasekhar, D. Bakowies, A. Glattli, P. H. Hünenberger, C. Pereira, and W. F. van Gunsteren, "Molecular dynamics simulation of lipid bilayers with GROMOS96: application of surface tension," *Mol. Simulation*, vol. 31, pp. 543–548, 2005.
- [207] S. J. Marrink, E. Lindahl, O. Edholm, and A. E. Mark, "Simulation of the spontaneous aggregation of phospholipids into bilayers," *J. Am. Chem. Soc.*, vol. 123, pp. 8638–8639, 2001.
- [208] Y. Gambin, R. Lopez-Esparza, M. Reffay, E. Siernecki, N. S. Gov, M. Genest, R. S. Hodges, and W. Urbach, "Lateral mobility of proteins in liquid membranes revisited," *Proc. Nat. Acad. Sci. USA*, vol. 103, pp. 2098–2102, 2006.
- [209] H. Nagase, H. Ueda, and M. Nakagaki, "Effect of water on lamellar structure of DPPC / sugar systems," *Biochim. Biophys. Acta*, vol. 1328, pp. 197–206, 1997.

- [210] W. L. Vaz, J. Stumpel, D. Hallmann, A. Gambacorta, and M. de Rosa, "Bounding fluid viscosity and translational diffusion in a fluid lipid bi-layer," *Eur. Biophys. J.*, vol. 15, pp. 111–115, 1987.
- [211] M. Ollmann, A. Robitzki, G. Schwarzmann, and H. J. Galla, "Minor effects of bulk viscosity on lipid translational diffusion measured by the excimer formation technique," *Eur. Biophys. J.*, vol. 16, pp. 109–112, 1988.
- [212] S. Cayley, B. A. Lewis, H. J. Guttman, and M. T. Record Jr., "Characterization of the cytoplasm of *Escherichia coli* K-12 as a function of external osmolarity. Implications for protein-DNA interactions *in vivo*," *J. Mol. Biol.*, vol. 222, pp. 281–300, 1991.
- [213] S. B. Zimmerman and S. O. Trach, "Estimation of macromolecule concentrations and excluded volume effects for the cytoplasm of *Escherichia coli*," *J. Mol. Biol.*, vol. 222, pp. 599–620, 1991.
- [214] S. Cayley and M. T. Record Jr., "Large changes in cytoplasmic biopolymer concentration with osmolality indicate that macromolecular crowding may regulate protein-DNA interactions and growth rate in osmotically stressed *Escherichia coli* K-12," *J. Mol. Recognit.*, vol. 17, pp. 488–496, 2004.
- [215] J. J. Spitzer and B. Poolman, "Electrochemical structure of the crowded cytoplasm," *Trends Biochem. Sci.*, vol. 30, pp. 536–541, 2005.
- [216] R. J. Ellis, "Macromolecular crowding: obvious but underappreciated," *Curr. Opin. Struct. Biol.*, vol. 11, pp. 114–119, 2001.
- [217] R. Swaminathan, C. P. Hoang, and A. S. Verkman, "Photo-bleaching recovery and anisotropy decay of green fluorescent protein GFP-S65T in solution and cells: cytoplasmic viscosity probed by green fluorescent protein translational and rotational diffusion," *Biophys. J.*, vol. 72, pp. 1900–1907, 1997.
- [218] M. B. Elowitz, M. G. Surette, P. E. Wolf, J. B. Stock, and S. Leibler, "Protein mobility in the cytoplasm of *Escherichia coli*," *J. Bacteriol.*, vol. 181, pp. 197–203, 1999.
- [219] C. W. Mullineaux, A. Nenninger, N. Ray, and C. Robinson, "Diffusion of green fluorescent protein in three cell environments in *Escherichia coli*," *J. Bacteriol.*, vol. 188, pp. 3442–3448, 2006.

- [220] M. C. Konopka, I. A. Shkel, S. Cayley, M. T. Record, and J. C. Weisshaar, "Crowding and confinement effects on protein diffusion *in vivo*," *J. Bacteriol.*, vol. 188, pp. 6115–6123, 2006.
- [221] A. Delon, Y. Usson, J. Derouard, T. Biben, and C. Souchier, "Continuous photo-bleaching in vesicles and living cells: a measure of diffusion and compartmentation," *Biophys. J.*, vol. 90, pp. 2548–2562, 2006.
- [222] M. J. Casadaban and S. N. Cohen, "Analysis of gene control signals by DNA fusion and cloning in *Escherichia coli*," *J. Mol. Biol.*, vol. 138, pp. 179–207, 1980.
- [223] R. S. Cormack and I. E. Somssich, "Cloning of PCR products via the green fluorescent protein," *Tech. Tips Online*, vol. 1, p. T01107, 1997.
- [224] A. Cramer, E. A. Whitehorn, E. Tate, and W. P. Stemmer, "Improved green fluorescent protein by molecular evolution using DNA shuffling," *Nat. Biotechnol.*, vol. 14, pp. 315–319, 1996.
- [225] S. Pautot, B. J. Frisken, and D. A. Weitz, "Engineering asymmetric vesicles," *Proc. Nat. Acad. Sci. USA*, vol. 100, pp. 10718–10721, 2003.
- [226] V. Noireaux and A. Libchaber, "A vesicle bioreactor as a step toward an artificial cell assembly," *Proc. Nat. Acad. Sci. USA*, vol. 101, pp. 17669–17674, 2004.
- [227] C. Eberhardt, L. Kuerschner, and D. S. Weiss, "Probing the catalytic activity of a cell division-specific transpeptidase *in vivo* with beta-lactams," *J. Bacteriol.*, vol. 185, pp. 3726–3734, 2003.
- [228] A. Ishihara, J. E. Segall, S. M. Block, and H. C. Berg, "Coordination of flagella on filamentous cells of *Escherichia coli*," *J. Bacteriol.*, vol. 155, pp. 228–237, 1983.
- [229] J. Starka, "Formation et stabilité osmotique des formes filamenteuses d'*Escherichia coli* induites par la pénicilline," *Ann. Inst. Pasteur (Paris)*, vol. 121, pp. 149–159, 1971.
- [230] E. Fischer, "Osmolability of *Escherichia coli* and modification of [125I]ampicillin-binding by competence induction for uptake of transforming DNA," *Arch. Microbiol.*, vol. 153, pp. 43–46, 1989.

- [231] R. D. Sleator and C. Hill, "Bacterial osmoadaptation: the role of osmolytes in bacterial stress and virulence," *FEMS Microbiol. Rev.*, vol. 26, pp. 49–71, 2002.
- [232] J. M. Wood, "Osmosensing by bacteria: signals and membrane-based sensors," *Microbiol. Mol. Biol. Rev.*, vol. 63, pp. 230–262, 1999.
- [233] I. Poirier, P.-A. Maréchal, C. Evrard, and P. Gervais, "*Escherichia coli* and *Lactobacillus plantarum* responses to osmotic stress," *Appl. Microbiol. Biotechnol.*, vol. 50, pp. 704–709, 1998.
- [234] R. Metzler and J. Klafter, "When translocation dynamics becomes anomalous," *Biophys. J.*, vol. 85, pp. 2776–2779, 2003.
- [235] M. Weiss, M. Elsner, F. Kartberg, and T. Nilsson, "Anomalous subdiffusion is a measure for cytoplasmic crowding in living cells," *Biophys. J.*, vol. 87, pp. 3518–3524, 2004.
- [236] I. M. Tolić-Nørrelykke, E.-L. Munteanu, G. Thon, L. Oddershede, and K. Berg-Sørensen, "Anomalous diffusion in living yeast cells," *Phys. Rev. Lett.*, vol. 93, pp. 78–102, 2004.
- [237] I. Golding and E. C. Cox, "Physical nature of bacterial cytoplasm," *Phys. Rev. Lett.*, vol. 96, pp. 98–102, 2006.
- [238] E. O. Potma, W. P. de Boeij, L. Bosgraaf, J. Roelofs, P. J. van Haastert, and D. A. Wiersma, "Reduced protein diffusion rate by cytoskeleton in vegetative and polarized *dictyostelium* cells," *Biophys. J.*, vol. 81, pp. 2010–2019, 2001.
- [239] P. Cluzel, M. Surette, and S. Leibler, "An ultrasensitive bacterial motor revealed by monitoring signaling proteins in single cells," *Science*, vol. 287, pp. 1652–1655, 2000.
- [240] T. van der Heide, M. C. Stuart, and B. Poolman, "On the osmotic signal and osmosensing mechanism of an ABC transport system for glycine betaine," *EMBO J.*, vol. 20, pp. 7022–7032, 2001.
- [241] D. Lu, P. Grayson, and K. Schulten, "Glycerol conductance and physical asymmetry of the *Escherichia coli* glycerol facilitator GlpF," *Biophys. J.*, vol. 85, pp. 2977–2987, 2003.
- [242] S. Cayley and M. T. Record Jr., "Roles of cytoplasmic osmolytes, water, and crowding in the response of *Escherichia coli* to osmotic stress: biophysical basis

- of osmoprotection by glycine betaine,” *Biochemistry*, vol. 42, pp. 12596–12609, 2003.
- [243] U. Dinnbier, E. Limpinsel, R. Schmid, and E. P. Bakker, “Transient accumulation of potassium glutamate and its replacement by trehalose during adaptation of growing cells of *Escherichia coli* K-12 to elevated sodium chloride concentrations,” *Arch. Microbiol.*, vol. 150, pp. 348–357, 1988.
- [244] A. Partikian, B. P. Ölveczky, R. Swaminathan, Y. Li, and A. S. Verkman, “Rapid diffusion of green fluorescent protein in the mitochondrial matrix,” *J. Cell Biol.*, vol. 140, pp. 821–829, 1998.
- [245] P. M. Haggie and A. S. Verkman, “Diffusion of tricarboxylic acid cycle enzymes in the mitochondrial matrix *in vivo*. Evidence for restricted mobility of a multienzyme complex,” *J. Biol. Chem.*, vol. 277, pp. 40782–40788, 2002.
- [246] B. P. Ölveczky and A. S. Verkman, “Monte carlo analysis of obstructed diffusion in three dimensions: application to molecular diffusion in organelles,” *Biophys. J.*, vol. 74, pp. 2722–2730, 1998.
- [247] Z. Gitai, “The new bacterial cell biology: moving parts and subcellular architecture,” *Cell*, vol. 120, pp. 577–586, 2005.
- [248] S. B. Zimmerman, “Shape and compaction of *Escherichia coli* nucleoids,” *J. Struct. Biol.*, vol. 156, pp. 255–261, 2006.
- [249] H. P. Kao, J. R. Abney, and A. S. Verkman, “Determinants of the translational mobility of a small solute in cell cytoplasm,” *J. Cell Biol.*, vol. 120, pp. 175–184, 1993.
- [250] G. S. Waldo, B. M. Standish, J. Berendzen, and T. C. Terwilliger, “Rapid protein-folding assay using green fluorescent protein,” *Nat. Biotechnol.*, vol. 17, pp. 691–695, 1999.
- [251] D. L. Bentley, “Rules of engagement: co-transcriptional recruitment of pre-mRNA processing factors,” *Curr. Opin. Cell Biol.*, vol. 17, pp. 251–256, 2005.
- [252] A. Köhler and E. Hurt, “Exporting RNA from the nucleus to the cytoplasm,” *Nat. Rev. Mol. Cell Biol.*, vol. 8, pp. 761–773, 2007.
- [253] M. Stewart, “Ratcheting mRNA out of the nucleus,” *Mol. Cell.*, vol. 25, pp. 327–330, 2007.

- [254] E. Lécuyer, H. Yoshida, N. Parthasarathy, C. Alm, T. Babak, T. Cerovina, T. R. Hughes, P. Tomancak, and H. M. Krause, "Global analysis of mRNA localization reveals a prominent role in organizing cellular architecture and function," *Cell*, vol. 131, pp. 174–187, 2007.
- [255] O. Isken and L. E. Maquat, "Quality control of eukaryotic mRNA: safeguarding cells from abnormal mRNA function," *Genes Dev.*, vol. 21, pp. 1833–1856, 2007.
- [256] Y. S. Huang and J. D. Richter, "Regulation of local mRNA translation," *Curr. Opin. Cell. Biol.*, vol. 16, pp. 308–313, 2004.
- [257] M. Müller, A. Heuck, and D. Niessing, "Directional mRNA transport in eukaryotes: lessons from yeast," *Cell. Mol. Life Sci.*, vol. 64, pp. 171–180, 2007.
- [258] I. G. Macara, "Transport into and out of the nucleus," *Microbiol. Mol. Biol. Rev.*, vol. 65, pp. 570–594, 2001.
- [259] D. C. Lee and J. D. Aitchison, "Kap104p-mediated nuclear import. Nuclear localization signals in mRNA-binding proteins and the role of Ran and RNA," *J. Biol. Chem.*, vol. 274, pp. 29031–29037, 1999.
- [260] J. T. Anderson, S. M. Wilson, K. V. Datar, and M. S. Swanson, "Nab2: a yeast nuclear polyadenylated RNA-binding protein essential for cell viability," *Mol. Cell. Biol.*, vol. 13, pp. 2730–2741, 1993.
- [261] R. E. Hector, K. R. Nykamp, S. Dheur, J. T. Anderson, P. J. Non, C. R. Urbinati, S. M. Wilson, L. Minvielle-Sebastia, and M. S. Swanson, "Dual requirement for yeast hnRNP Nab2p in mRNA poly(A) tail length control and nuclear export," *EMBO J.*, vol. 21, pp. 1800–1810, 2002.
- [262] S. M. Kelly, S. A. Pabit, C. M. Kitchen, P. Guo, K. A. Marfatia, T. J. Murphy, A. H. Corbett, and K. M. Berland, "Recognition of polyadenosine RNA by zinc finger proteins," *Proc. Nat. Acad. Sci. USA*, vol. 104, pp. 12306–12311, 2007.
- [263] E. J. Tran, Y. Zhou, A. H. Corbett, and S. R. Wenthe, "The DEAD-box protein Dbp5 controls mRNA export by triggering specific RNA:protein remodeling events," *Mol. Cell*, vol. 28, pp. 850–859, 2007.
- [264] D. M. Green, K. A. Marfatia, E. B. Crafton, X. Zhang, X. Cheng, and A. H. Corbett, "Nab2p is required for poly(A) RNA export in *Saccharomyces cerevisiae* and is regulated by arginine methylation via Hmt1p," *J. Biol. Chem.*, vol. 277, pp. 7752–7760, 2002.

- [265] K. K. Guisbert, K. Duncan, H. Li, and C. Guthrie, "Functional specificity of shuttling hnRNPs revealed by genome-wide analysis of their RNA binding profiles," *RNA*, vol. 11, pp. 383–393, 2005.
- [266] K. Duncan, J. G. Umen, and C. Guthrie, "A putative ubiquitin ligase required for efficient mRNA export differentially affects hnRNP transport," *Curr. Biol.*, vol. 10, pp. 687–696, 2000.
- [267] M. M. Kessler, M. F. Henry, E. Shen, J. Zhao, S. Gross, P. A. Silver, and C. L. Moore, "Hrp1, a sequence-specific RNA-binding protein that shuttles between the nucleus and the cytoplasm, is required for mRNA 3'-end formation in yeast," *Genes Dev.*, vol. 11, pp. 2545–2556, 1997.
- [268] M. F. Henry, D. Mandel, V. Routson, and P. A. Henry, "The yeast hnRNP-like protein Hrp1/Nab4 accumulates in the cytoplasm after hyperosmotic stress: a novel Fps1-dependent response," *Mol. Biol. Cell*, vol. 14, pp. 3929–3941, 2003.
- [269] L. H. Apponi, S. M. Kelly, M. T. Harreman, A. N. Lehner, A. H. Corbett, and S. R. Valentini, "An interaction between two RNA binding proteins, Nab2 and Pub1, links mRNA processing/export and mRNA stability," *Mol. Cell Biol.*, vol. 27, pp. 6569–6579, 2007.
- [270] M. Windgassen, D. Sturm, I. J. Cajigas, C. I. Gonzalez, M. Seedorf, H. Bastians, and H. Krebber, "Yeast shuttling SR proteins Npl3p, Gbp2p, and Hrb1p are part of the translating mRNPs, and Npl3p can function as a translational repressor," *Mol. Cell Biol.*, vol. 24, pp. 10479–10491, 2004.
- [271] J. D. Aitchison, G. Blobel, and M. P. Rout, "Kap104p: a karyopherin involved in the nuclear transport of messenger RNA binding proteins," *Science*, vol. 274, pp. 624–627, 1996.
- [272] C. I. González, M. J. Ruiz-Echevarra, S. Vasudevan, M. F. Henry, and S. W. Peltz, "The yeast hnRNP-like protein Hrp1/Nab4 marks a transcript for nonsense-mediated mRNA decay," *Mol. Cell*, vol. 5, pp. 489–499, 2000.
- [273] W. K. Huh, J. V. Falvo, L. C. Gerke, A. S. Carroll, R. W. Howson, J. S. Weissman, and O. K. O'Shea, "Global analysis of protein localization in budding yeast," *Nature*, vol. 425, pp. 686–691, 2003.
- [274] E. A. Winzeler, D. D. Shoemaker, A. Astromoff, H. Liang, K. Anderson, B. Andre, R. Bangham, R. Benito, J. D. Boeke, H. Bussey, A. M. Chu, C. Connelly, K. Davis, F. Dietrich, S. W. Dow, M. E. Bakkoury, F. Foury, S. H.

- Friend, E. Gentalen, G. Giaever, J. H. Hegemann, T. Jones, M. Laub, H. Liao, N. Liebundguth, D. J. Lockhart, A. Lucau-Danila, M. Lussier, N. M'Rabet, P. Menard, M. Mittmann, C. Pai, C. Rebischung, J. L. Revuelta, L. Riles, C. J. Roberts, P. Ross-MacDonald, B. Scherens, M. Snyder, S. Sookhai-Mahadeo, R. K. Storms, S. Véronneau, M. Voet, G. Volckaert, T. R. Ward, R. Wysocki, G. S. Yen, K. Yu, K. Zimmermann, P. Philippsen, M. Johnston, and R. W. Davis, "Functional characterization of the *S. cerevisiae* genome by gene deletion and parallel analysis," *Science*, vol. 285, pp. 901–906, 1999.
- [275] B. L. Timney, J. Tetenbaum-Novatt, D. S. Agate, R. Williams, W. Zhang, B. T. Chait, and M. P. Rout, "Simple kinetic relationships and nonspecific competition govern nuclear import rates *in vivo*," *Cell Biol.*, vol. 175, pp. 579–593, 2006.
- [276] B. R. Martin, B. N. Giepmans, B. R. Adams, and R. Y. Tsien, "Mammalian cell-based optimization of the biarsenical-binding tetracysteine motif for improved fluorescence and affinity," *Nat. Biotechnol.*, vol. 23, pp. 1308–1314, 2005.
- [277] R. K. Niedenthal, L. Riles, M. Johnston, and J. H. Hegemann, "Green fluorescent protein as a marker for gene expression and subcellular localization in budding yeast," *Yeast*, vol. 12, pp. 773–786, 1996.
- [278] C. B. Brachmann, A. Davies, G. J. Cost, E. Caputo, J. Li, P. Hieter, and J. D. Boeke, "Designer deletion strains derived from *Saccharomyces cerevisiae* S288C: a useful set of strains and plasmids for PCR-mediated gene disruption and other applications," *Yeast*, vol. 14, pp. 115–132, 1998.
- [279] M. Winey, D. Yarar, T. H. Giddings Jr., and D. N. Mastronarde, "Nuclear pore complex number and distribution throughout the *Saccharomyces cerevisiae* cell cycle by three-dimensional reconstruction from electron micrographs of nuclear envelopes," *Mol. Biol. Cell.*, vol. 8, pp. 2119–2132, 1997.
- [280] R. M. Long, D. J. Elliott, F. Stutz, M. Rosbash, and R. H. Singer, "Spatial consequences of defective processing of specific yeast mRNAs revealed by fluorescent *in situ* hybridization," *RNA*, vol. 1, pp. 1071–1078, 1995.
- [281] A. J. Rodriguez, S. M. Shenoy, R. H. Singer, and J. Condeelis, "Visualization of mRNA translation in living cells," *J. Cell Biol.*, vol. 175, pp. 67–76, 2006.

- [282] M. Andresen, R. Schmitz-Salue, and S. Jakobs, "Short tetracysteine tags to beta-tubulin demonstrate the significance of small labels for live cell imaging," *Mol. Biol. Cell*, vol. 15, pp. 5616–5622, 2004.
- [283] S. R. Adams, R. E. Campbell, L. A. Gross, B. R. Martin, G. K. Walkup, Y. Yao, J. Llopis, and R. Y. Tsien, "New biarsenical ligands and tetracysteine motifs for protein labeling *in vitro* and *in vivo*: synthesis and biological applications," *J. Am. Chem. Soc.*, vol. 124, pp. 6063–6076, 2002.
- [284] J. Lippincott-Schwartz, E. Snapp, and A. Kenworthy, "Studying protein dynamics in living cells," *Nat. Rev.*, vol. 2, pp. 444–456, 2001.
- [285] W. Yang, J. Gelles, and S. M. Musser, "Imaging of single-molecule translocation through nuclear pore complexes," *Proc. Nat. Acad. Sci. USA*, vol. 101, pp. 12887–12892, 2004.
- [286] R. M. Long, R. H. Singer, X. Meng, I. Gonzalez, K. Nasmyth, and R. P. Jansen, "Mating type switching in yeast controlled by asymmetric localization of *ASH1* mRNA," *Science*, vol. 277, pp. 383–387, 1997.
- [287] P. A. Takizawa, A. Sil, J. R. Swedlow, I. Herskowitz, and R. D. Vale, "Actin-dependent localization of an RNA encoding a cell-fate determinant in yeast," *Nature*, vol. 389, pp. 90–93, 1997.
- [288] O. Keminer, J. P. Siebrasse, K. Zerf, and R. Peters, "Optical recording of signal-mediated protein transport through single nuclear pore complexes," *Proc. Nat. Acad. Sci. USA*, vol. 96, pp. 11842–11847, 1999.
- [289] T. Gross, A. Siepmann, D. Sturm, M. Windgassen, J. J. Scarcelli, M. Sedorf, C. N. Cole, and H. Krebber, "The DEAD-box RNA helicase Dbp5 functions in translation termination," *Science*, vol. 315, pp. 646–649, 2007.
- [290] K. Asakawa and A. Toh-e, "A defect of Kap104 alleviates the requirement of mitotic exit network gene functions in *Saccharomyces cerevisiae*," *Genetics*, vol. 162, pp. 1545–1556, 2002.
- [291] C. P. Lusk, D. D. Waller, T. Makhnevych, A. Dienemann, M. Whiteway, D. Y. Thomas, and R. W. Wozniak, "Nup53p is a target of two mitotic kinases, Cdk1p and Hrr25p," *Traffic*, vol. 8, pp. 647–660, 2007.
- [292] S. W. Jones, R. Christison, K. Bundell, C. J. Voyce, S. M. V. Brockbank, P. Newham, and M. A. Lindsay, "Characterisation of cell-penetrating peptide-mediated peptide delivery," *Br. J. Pharmacol.*, vol. 145, pp. 1093–1102, 2005.

- [293] H. Brooks, B. Lebleu, and E. Vives, "Tat peptide-mediated cellular delivery: back to basics," *Adv. Drug Del. Rev.*, vol. 57, pp. 559–577, 2005.
- [294] M. F. Engel, L. Khemtémourian, C. C. Kleijer, H. J. Meeldijk, J. Jacobs, A. J. Verkleij, B. De Kruijff, J. A. Killian, and J. W. Höppener, "Membrane damage by human islet amyloid polypeptide through fibril growth at the membrane," *Proc. Nat. Acad. Sci. USA*, vol. 105, pp. 6033–6038, 2008.
- [295] M. Frick, K. Schmidt, and B. J. Nichols, "Modulation of lateral diffusion in the plasma membrane by protein density," *Curr. Biol.*, vol. 17, pp. 462–467, 2007.
- [296] J. Ries and P. Schwille, "Studying slow membrane dynamics with continuous wave scanning fluorescence correlation spectroscopy," *Biophys. J.*, vol. 91, pp. 1915–1924, 2006.
- [297] Q. Q. Ruan, M. A. Cheng, M. Levi, E. Gratton, and W. W. Mantulin, "Spatial-temporal studies of membrane dynamics: scanning fluorescence correlation spectroscopy (SFCS)," *Biophys. J.*, vol. 87, pp. 1260–1267, 2004.
- [298] C. M. Brown, R. B. Dalal, B. Hebert, M. A. Digman, A. R. Horwitz, and E. Gratton, "Raster image correlation spectroscopy (RICS) for measuring fast protein dynamics and concentrations with a commercial laser scanning confocal microscope," *J. Microsc.*, vol. 229, pp. 78–91, 2008.
- [299] G. Guigas and M. Weiss, "Sampling the cell with anomalous diffusion - the discovery of slowness," *Biophys. J.*, vol. 94, pp. 90–94, 2008.
- [300] S. W. Hell, "Toward fluorescence nanoscopy," *Nat. Biotechnol.*, vol. 11, pp. 1347–1355, 2003.
- [301] J. H. Rice, "Beyond the diffraction-limit: far-field fluorescence imaging with ultrahigh resolution," *Mol. Biosyst.*, vol. 3, pp. 781–793, 2007.
- [302] B. Harke, J. Keller, C. K. Ullal, V. Westphal, A. Schönle, and S. W. Hell, "Resolution scaling in STED microscopy," *Opt. Exp.*, vol. 16, pp. 4154–4162, 2008.
- [303] K. Peneva, G. Mihov, F. Nolde, S. Rocha, J. I. Hotta, K. Braeckmans, J. Hofkens, H. Uji-I, A. Herrmann, and K. Müllen, "Water-soluble monofunctional perylene and terylene dyes: powerful labels for single-enzyme tracking," *Angew. Chem. Int. Ed. Engl.*, vol. 47, pp. 3372–3375, 2008.

- [304] N. C. Shaner, R. E. Campbell, P. A. Steinbach, B. N. G. Giepmans, A. E. Palmer, and R. Y. Tsien, "Improved monomeric red, orange and yellow fluorescent proteins derived from *Discosoma* sp. red fluorescent protein," *Nat. Biotechnol.*, vol. 22, pp. 1567–1572, 2004.
- [305] A. Tirat, F. Freuler, T. Stettler, L. M. Mayr, and L. Leder, "Evaluation of two novel tag-based labelling technologies for site-specific modification of proteins," *Int. J. Biol. Macromol.*, vol. 39, pp. 66–67, 2006.
- [306] D. C. Nga, H. Tamura, A. Tokuda, T. Yamamotoa, M. Matsuo, M. Nunoshita, Y. Ishikawa, S. Shiosaka, and J. Ohta, "Real time *in vivo* imaging and measurement of serine protease activity in the mouse hippocampus using a dedicated complementary metal-oxide semiconductor imaging device," *J. Neurosci. Methods*, vol. 156, pp. 23–30, 2006.