MEMPHYS – Center for Biomembrane Physics

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MEMPHYS - Center for Biomembrane Physics is an interdisciplinary research center funded by the Danish National Research Foundation for the period 2001-2011. The Center is concerned with parallel experimental, computational, and theoretical research within the broad fields of physics and physical chemistry of soft interfaces and biological membranes. The focus is on developing molecular descriptions of the physical and physico-chemical properties of membrane systems and investigating how these properties control membrane function [1].

Our roots and research strategy

The scientists who founded MEMPHYS had their feet solidly planted in the physical sciences, specifically statistical physics, thermodynamics, computational physics, physical chemistry, biophysical chemistry, biological chemistry, soft and hard condensed matter physics, and the physics of complex systems. These disciplins and their associated methodologies are rather robust and carry easily over to be used in other fields, such as the life sciences. In particular they allow for massive cross-disciplinarity, spanning physics, chemistry, and biology, as well as spanning the three pillars of modern natural science: 1) experiment, 2) theory, and 3) modeling, simulational, and computational science. The main research strategy adopted by MEMPHYS is, in the *same* center and under the *same* roof, to exploit the full potential of building on all three pillars.

Table 1. Some systems and phenomena studied at MEMPHYS

Lipids, proteins, peptides, nucleic acids, carbohydrates
(Bio)polymers, poly-electrolytes
Lipid monolayers
Lipid bilayers (vesicles, liposomes, supported bilayers)
Biological membranes
Cell organelles, nuclei, and whole cells
Interactions with enzymes, proteins, peptides, nucleic acids, drugs, alcohols, sterols, antibiotics, as well as other compounds that are active in/at membranes
DNA and stochastic processes
Ligand-receptor interactions
Lung surfactants
Skin and dermal barrier

The areas of research at MEMPHYS cover lipid monolayers, bilayers, and biological membranes as well as interactions of these systems with enzymes, polynucleotides, proteins, peptides, drugs, antibiotics, alcohols, sterols, as well as other compounds that are active in membranes (**Table 1**). The Center exploits a multitude of theoretical and experimental methods and techniques (**Table 2**). The Center is strongly engaged in national and international collaborations and is furthermore committed to strengthening the training of young researchers in molecular biophysics and biophysical chemistry. Results of the research are applied within the pharmaceutical and biomedical area as well as within nano-science, nano-biotechnology, and food science where the traditional borders between physics, chemistry, and biology tend to vanish. Members of MEMPHYS have a range of collaborative projects with industry and companies within the sectors of biomedicine, nano- and biotechnology, food technology, scientific instrumention, and software development.

The Center has three locations, with the main node at SDU and smaller nodes at DTU and RUC. The Center members work across traditional scientific boundaries and span existing departmental borders. In addition to basic science and curiousity-driven research, Center members are occupied with training scientists, in particular PhDs, who get jobs in academia, industry, companies, teaching institutions, hospitals, technological institutes, as well as communication and administration units.

Research themes and the model paradigm

The research at MEMPHYS is loosely organized in a number of themes that are dynamic and change over time (**Table 3**). In addition they are highly interrelated.

With roots in the physical sciences it is natural to approach the structure and function of biological membranes via a range of well-defined models; experimental, theoretical, as well as computational models [Mouritsen, *Cold Spring Harb. Perspect. Biol.* 3, 33-47, 2011]. A 'model' is the tie that connects theory and experiment and it is a necessary prerequisite for doing truly quantitative membrane research. A model and its associated hypotheses constitute the starting point for any experiment, and a model is furthermore necessary for proper interpretation of the experimental data. A

model is a dynamicentity that changes as the interplay between theory, experiment, and simulation evolves.

Table 2. Research expertise, methods and facilities at MEMPHYS

Parallel experimental, theoretical, and simulational research
Physics and physical chemistry of soft interfaces and biological membranes
Molecular modeling and computer simulation techniques (MC, MD, DPD)
Thermodynamic measurements (DSC, ITC calorimetry, vapor pressure)
Monolayer techniques (Langmuir, Langmuir Blodgett)
Biophotonics, fluorescence spectroscopy, (spinning-disc) confocal microscopy, fluorescence microscopy (multi-photon excitation and coupling to micropipette aspiration/manipulation), TIRF, STED, FLIM, SHGM
Biological imaging by quantum dots, single-particle tracking
Magnetic resonance spectroscopy (EPR)
Neutron reflectometry, X-ray, and neutron diffraction
Micromechanics and micro-manipulation (vesicle-fluctuation analysis and micropipette aspiration/manipulation)
Ultra-sensitive surface-probe techniques and single-molecule detection methods, such as atomic force microscopy, imaging ellipsometry, and bioprobe force spectroscopy
GMO Class 1 cell laboratory
Extensive access to super-cluster computational installations under DCSC-Danish Center for Scientific Computing at SDU

In **Fig. 1** is given a gallery of images characterizing some of the membrane models that are being worked with in MEMPHYS. The following is a brief tour through this gallery that reflects some of the work that has been carried out at MEMPHYS over the last few years. Some representative references are given; for a full list of references, see [2] where a fuller account of the MEMPHYS research is also to be found.

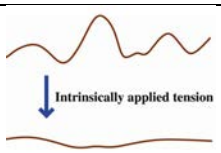
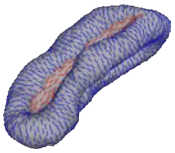
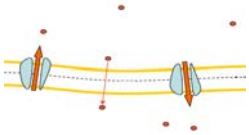
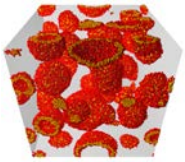
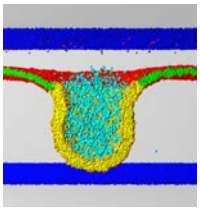
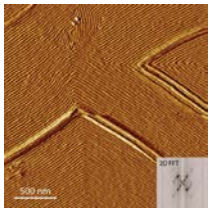
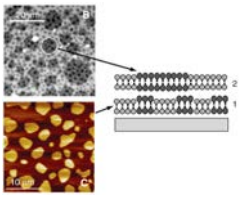
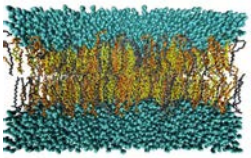
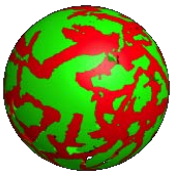
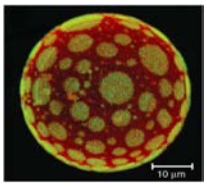
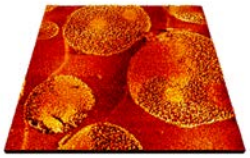
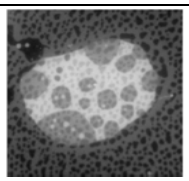
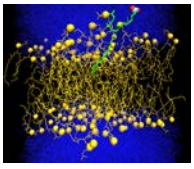
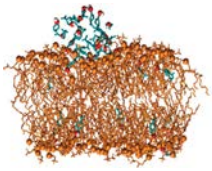
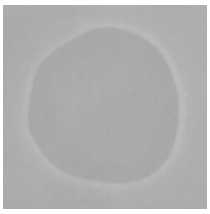
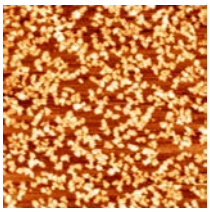
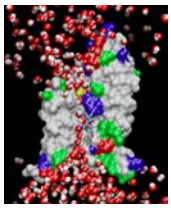
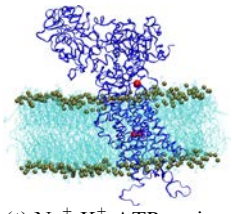
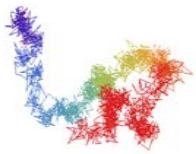
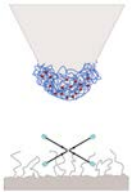
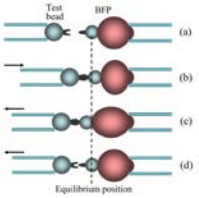
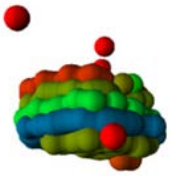
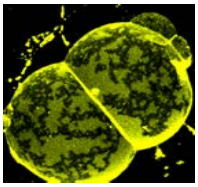
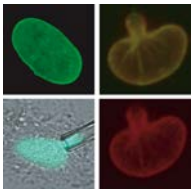
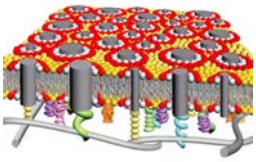
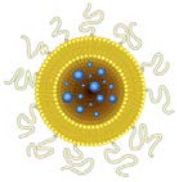
Membranes as thin sheets of self-assembled molecules

A simple model of a membrane anticipates its effective two-dimensional geometric character as a thin sheet (**a**) embedded in 3-d space and associated with mechanical and topological properties. The fluctuating character of the sheet is controlled by temperature, bending and area-compression modules, as well as tension [Lomholt, Loubet, Ipsen, *Phys. Rev. E* 83, 011913 (2011]. A closed sheet and its statistical thermodynamics can be modeled by a fluid, randomly triangulated network (**b**). In-plane ordering and a complex morphology can arise in the case of different types of membrane proteins with different preferred curvature, which may lead to to phase separate in the plane of the membrane [Ramakrishnan, Kumar, Ipsen, *Phys. Rev E* 81, 041922, 2010]. In the case where protein activity is coupled to the membrane sheet (**c**), an effective renormalization (softening) of the bending rigidity sets in which has observed experimentally in giant unilamellar liposomes reconstituted with active Na^+-K^+-ATPase [Bouvrais, *PhD Thesis*, SDU, 2011].

Table 3. Main research themes at MEMPHYS

Basic physical properties of lipid membranes and monolayers
Single-molecule biophysics and hydrophobic and solvent-mediated forces
Physics of biopolymers and fibers and their interaction with membranes
Non-equilibrium properties of membranes
Novel membrane models: supported membranes and reconstituted vesicles
Liposomes and drug delivery
Interaction of antibiotics and peptides with membranes and bacteria
Solutes and their interaction with membranes and proteins
The effects of sterols on membrane structure and function
Lateral membrane structure: lipid domains and functional rafts
Lipid-protein interactions and membrane transport
Glycosylated membranes
Structure-function relationship of enzymes
Complex membranes
Instrumentation and software development
Biophysics and didactics

Membranes owe their existence and peculiar materials properties to the fact that they are macromolecular assemblies of amphiphilic molecules in water. Using specially designed software developed to be executed at a parallel architecture of computers it has become possible to model and simulate the self-assembly process of very large collections of molecules (e.g., ~100.000 amphiphiles in ~5 mio water molecules) over sufficiently long time scales (microseconds) to monitor the self-asembly from lipid monomers in aqueous solution to ensembles of vesicles (**d**) [Shillcock, *Langmuir*, dx.doi.org/10.1021/la2033803, 2012]. The simulations proceed by dissipative particle dynamics and coarse-grained models which also allow for the study of complex processes involving membrane curvature, e.g., in the form of cap formation due to local phase separation of different lipid species (**e**) [Laradji, Kumar, *Phys. Rev. E* 73, 040901, 2006]], or fusion of vesicles with target membra-

 (**a**) Fluctuating membrane under intrinsic tension.	 (**b**) Curvature-induced protein segregation.	 (**c**) Membrane with active proteins.	 (**d**) Self-assembly of liposomes in water.
 (**e**) Phase-separation-induced butting on a vesicle.	 (**f**) Vesicle-membrane fusion.	 (**g**) Ripple-phase structure in a binary lipid bilayer.	 (**h**) Solid-supported ternary-lipid bilayers.
 (**i**) Raft formation in model membranes.	 (**j**) Domains in giant unilamellar vesicles.	 (**k**) Texture of solid lipid domains in membranes.	 (**l**) Domains in pulmonary surfactant membranes.
 (**m**) Pulmonary surfactant membranes.	 (**n**) Membrane remodeling by lipases.	 (**o**) Oxidized lipid protrusion in membranes.	 (**p**) Perillyl alcohol binding to membranes.
 (**q**) Fluctuating vesicle with antimicrobial peptides.	 (**r**) Aquaporins in a supported lipid bilayer.	 (**s**) Water transport in aquaporin in a bilayer.	 (**t**) Na^+-K^+-ATPase in a lipid-bilayer membrane.
 (**u**) Single-particle trace of a single lipid in a live cell.	 (**v**) Probing ligand-receptor interactions.	 (**x**) Soft-probe force spectroscopy.	 (**y**) Model of Golgi-complex formation.
 (**z**) Model of skin-lipid membranes.	 (**æ**) Pipette aspiration of a nuclear membrane.	 (**ø**) Model of a complex membrane with rafts.	 (**å**) Liposome as drug-delivery vehicle.

Figur 1. Gallery of some membrane models studied at MEMPHYS.

nes, modeling the fusion of synaptic vesicles loaded with neurotransmitters (**f**) or translocation of nano-particles across a membrane [Shillcock, *HFSP J*. 2, 1-6, 2008].

Lateral membrane structure: domains and rafts

One of the absolute key issues in membrane science in recent decades has been the unraveling of the lateral structure of biological membranes and how this lateral structure controls function. A very convenient model of a membrane is a monolayer positioned at the air-water interface, a so-called Langmuir film, whose thermodynamics and thermomechanics can be studied in a Langmuir trough and whose lateral structure can be investigated by fluorescence microscopy and Brewster-angle microscopy. Monolayers can be transferred to solid substrates to form Langmuir-Blodgett films. These films, which also can be formed by spin coating or by explosion and fusion of vesicles onto the substrate, are ideal models of freestanding bilayer membranes in water, e.g., under physiological conditions, and they can be studied in detail by fluorescence microscopy, ellipsometry, or atomic force microscopy. Double-supported bilayer assays provide for fully unperturbed distal bilayers and allow e.g., for the observation of the corrugated ripple structure of solid bilayers (**g**) [Bagatolli, Ipsen, Simonsen, Mouritsen, *Prog. Lip. Res.* 49, 378–389, 2010] or domain formation of ternary lipid mixtures with cholesterol (**h**). The latter mixture is the prototype of a model membrane exhibiting raft formation [Jensen, Morris, Simonsen, *Langmuir* 2007, 23, 8135-8141, 2007]. Raft formation in bilayers containing sphingomyelin and cholesterol has also been studied by molecular dynamics simulation techniques (**i**) [Niemela, Ollila, Hyvonen, Karttunen, Vattulainen, *PLoS Comp. Biol.* 3, e34, 2007].

The longstanding puzzle has been resolved regarding the relationship between thermodynamic phase equilibria in many-component lipid membranes and the observation of domain patterns at the surface of giant unilamellar vesicles (GUVs) (**j**). Stereological analysis of area fractions on the curved surface permits the examination of the thermodynamic lever rule and the results demonstrate that the coexistence of lipid domains corresponds to equilibrium thermodynamic phases, hence proving that GUVs are good models of lipid membranes [Fidorra, Garcia, Ipsen, Hartel, Bagatolli, *Biochim. Biophys. Acta* 1788, 2142–2149, 2009]. A more subtle aspect of lipid domain formation is the molecular orientational order inside the domains. The very first multi-photon polarization fluorescence microscopy study of planar lipid bilayers has demonstrated that the texture of gel domains in binary lipid membranes showing gel/liquid-disordered phase coexistence displays the rare hexatic order (**k**) [Bernchou, Brewer, Midtiby, Ipsen, Bagatolli, Simonsen, *J. Amer. Chem. Soc.* 131, 14130-14131, 2009].

Moving to more complex membranes, the pulmonary surfactant content of lung lining lavage extracts has been studied both in the form of giant vesicles (**l**) and as supported bilayers (**m**). These extracts contain various lipids, including cholesterol, as well as pulmonary surfactant proteins. The work shows for the first time the existence of liquid-ordered domains ('rafts') in cell membranes and that the proteins tend to localize in these domains [Bernardino de la Serna, Oradd, Bagatolli, Simonsen, Marsh, Lindblom, Perez-Gil, *Biophys. J.* 97, 1381-1389, 2009].

Membranes can be degraded and remodeled by the action of interfacially active enzymes like phospholipases, e.g. phospholipase A_2. The action of a lipase on a membrane can be monitored and charactericed quantitatively by time-series analysis of images recorded by atomic force microscopy or fluorescence microscopy (**n**) e.g., on ternary, double-supported bilayers of mixtures of cholesterol with lipids of high and low melting points ('raft mixture'). The analysis shows that activation by a uniform membrane in the liquid-disordered phase leads to nucleation and growth of liquid-ordered-like domains [Simonsen, *Biophys. J.* 94, 3966–3975, 2008].

Membranes interacting with 'foreign' molecules

A recurrent research theme at MEMPHYS has been studies of various compound ('foreign molecules') interacting with membranes, e.g., specific lipids, sterols, drugs, peptides, and bioactive molecules of various kinds. The strategy has been to study and characterize these interactions by measuring how the various compounds affect the termodynamic, thermomechanic, as well as physical properties of the membranes on scales from the individual molecule to the size of the membrane.

A particular example is membrane modification due to contamination by lipid oxidation products, a phenomenon of major importance for ageing of cells and various diseases. In oxidative environments, biomembranes contain oxidized lipids with short, polar acyl chains. Molecular Dynamics simulations have provided the first molecular evidence of the socalled extended lipid conformation in phospholipid membranes (**o**). The chain reversal of some oxidized lipids decorates the membrane interface with reactive, negatively charged functional groups. Such chain reversal is likely to exert a profound influence on the structure and dynamics of biological membranes and on membrane-associated biological processes [Khandelia, Mouritsen, *Biophys. J.* 96, 2734-2743, 2009].

Many bioactive natural compounds are supposed to exert their action, e.g., antimicrobial, on mem-

branes. An example is the biooxidation products from the *Perilla* plant that is widely used in traditional Asian medicine as well as for preservation of vegetables and fruits in the Japanese cuisine. Molecular Dynamics simulations together with thermodynamic and spectroscopic experiments have shown that at least some medicinal properties of the volatile *Perilla* extracts might arise from interactions with the lipid bilayer component of biological membranes (**p**) [Witzke, Duelund, Kongsted, Petersen, Mouritsen, Khandelia, *J. Phys. Chem. B* 114, 15825–15831, 2010].

The effects of membrane active compounds, e.g., antimicrobial peptides, can be monitored by vesicle-fluctuation analysis by which the contour of large unilamellar vesicles (**q**) are analysed by Fourier analysis in order to determine the bending modulus. As an example the natural peptide magainin has been found to soften the bilayer and induce holes facilitating ion leakage [Bouvrais, Méléard, Pott, Jensen, Brask, Ipsen, *Biophys. Chem.* 137, 7-12, 2008].

Membranes with integral proteins

Integral membrane proteins can be reconstituted into well-defined lipid bilayers and then be subjected to the same kind of biophysical investigations as simple lipid-bilayer membranes. An example is water-transporting aquaporins, e.g. from spinach, which can be imaged on a single-molecule level by atomic force microscopy (**r**), and the effect of the lipids on the protein conformation and stabilility can be assessed by circular dicroism [Plasencia, Survery, Ibragimova, Hansen, Nielsen, Kjellbom, Johanson, O. G. Mouritsen, *PLoS ONE* 6, e14674, 1-9, 2011]. Molecular Dynamics simulations in turn can help to reveal the transport properties of the channel and how they depend on the nature of the lipid matrix (**s**) [Khandelia, Jensen, Mouritsen, *J. Phys. Chem. B.* 16, 5239-5244, 2009].

Another important class of membrane proteins is Na^+-K^+-ATPase that is absolutely crucial for maintaining the proper balance of the small ions across all cell membranes. This ion pump actively exchanges three sodium ions for two potassium ions across the cell via what so far has been believed to be a single ion pathway. Molecular Dynamics simulations (**t**) of the wild-type enzyme and its C-terminal mutants embedded in a lipid bilayer have revealed an additional pathway leading to the ion-binding site, which spontaneously fills up with water molecules. Combined with electrophysiology experiments, the simulations suggest, for the first time, that an additional ion pathway from the C-terminus to the ion binding site exists in the Na^+-K^+-ATPase, through which protons can diffuse into the binding site to preserve the asymmetric stoichiometry of ion transport [Poulsen, Khandelia, Morth, Mouritsen, Jensen, Nissen, *Nature* 467, 99-102, 2010].

Complex membranes and cell membranes

Advanced biophotonics equipment developed and installed in the MEMPHYS laboratories has made it possible to image the tracking of individual molecules, such as appropriately fluorescently labeled proteins or lipids, in intact membranes of living cells, e.g. in a live mouse embryo fibroblast [Clausen, Lagerholm, *Curr. Protein Pept. Sci.* 12, 699-713, 2011]. The trajectory shown in (**u**) corresponds to a single lipid molecule (biotin-cap-DPPE) that has been labeled with a streptavidin-functionalized quantum dot (QD655) and imaged at 1700 Hz. From the traces the diffusion constant can be determined and indirect information can be obtained about the local structure of the environment in which the molecule diffuses. Similarly, techniques have been developed to image single-protein motion by labeling the proteins with quantum dots.

Single-molecule biophysical techniques can also be used to quantitatively measure the dynamic bonding strength and rupture force of individual ligand-receptor interactions. Two very different approaches have been implemented in the MEMPHYS laboratories. In the first approach the cantilever in an atomic force microscope measures the force spectrum. Using a properly functionalized tip of the cantilever it is possible to investigate the binding between a single receptor molecule that is bound to a surface and a single ligand molecule that is bound to the tip (**v**). Upon pulling on the bound complex, the dynamic binding strength of the ligand-receptor interaction can be investigated quantitatively in terms of a force-distance relationship. An example is the study of the binding between pulmonary surfactant protein D (SP-D), a collectin in the native immune system of the lung, and various sugars. The protein's ability to discriminate the binding to different sugars lies at the root of the molecular mechanism of the immune system. It has been found that SP-D binds strongest to D-mannose and weakest to maltose and D-galactose which may explain the ability of the protein to selectively identify bacterial surfaces [Thormann, Dreyer, Simonsen, Hansen, Holmskov, Hansen, Mouritsen, *Biochemistry* 46, 12231-12237 (2007].

The other technique, called bioprobe or soft-probe force spectroscopy, is a rare speciality and only practised in a few laboratories around the world. It is based on a principle where the inter-molecular force can be monitored by the distortion of a soft body, e.g., a swolled red blood cell or a liposome, attached to a fine glas capillary tube operated by a suction pressure (**x**). An example is the study of integrins that require divalent ions for ligand recognition. Mechanically enforced bond dissociation in the soft-force probe spectrometer has revealed a synergistic influence of Mn^{2+} and Mg^{2+} on the interaction between integrin $\alpha 7\beta 1$ and inva-

sin [Ligezowska, Boye, Eble, Hoffmann, Klösgen, Merkel, *J. Mol. Recognit.* 24, 715-723, 2011].

The Golgi apparatus is an example of an internal cell structure whose morphogenesis and compartmentalization is a consequence of a membrane-mediated non-equilibrium phenomenon that is not understood in detail. The Golgi apparatus is the central hub for protein sorting and lipid metabolism in the secretory pathway. A new coarse-grained computational model that captures key features of the dynamic morphogenesis (**y**) has related the experimentally observed Golgi phenotypes, the typical turnover times, and the size and number of cisternae to three basic, experimentally accessible quantities: the rates for material influx from the endoplasmic reticulum, and the anterograde and retrograde transport rates. Based on these results it has been proposed which molecular factors should be mutated in order to alter the organelle's phenotype and dynamics [Kühnle, Shillcock, Mouritsen, Weiss, *Biophys. J.* 98, 2839–2847, 2010].

The skin is a complex organ whose barrier properties derives from the peculiar structure of the *stratum corneum* whose structure in humans recently has been revealed by fluorescence microscopy techniques [Iwai, Han, Svensson, Öfverstedt, Anwar, Brewer, Bloksgaard, den Holländer, Laloeuf, Nosek, Masich, Bagatolli, Skoglund, Norlén. *J. Investig. Dermatol.* in press, 2012]. The dermal barrier capacity is a function of the physical state and the structural organization of the *stratum corneum* extracellular lipid matrix. This lipid matrix is essentially composed of very long-chain saturated ceramides, cholesterol, and free fatty acids. Diect visualization of hydrated bilayers of human skin *stratum corneum* lipids has shown these bilayers to exhibit a giant sponge-like morphology with dimensions corresponding to the global three-dimensional morphology of the *stratum corneum* extracellular space (**z**) [Plasencia-Gil, Norlén, Bagatolli, *Biophys. J.* 93, 3142-3155, 2007].

An even more complex biological membrane is the nuclear envelope membrane whose physical properties for the first time was investigated, using living human epithelial (HeLa) cell nuclei, by a combination of micropipette-aspiration studies (æ) and a theoretical analysis in terms of the elasticity of a two-dimensional body. By confocal microscopy and micromanipulation of the nuclear envelope in living cells and isolated nuclei it was found that the envelope undergoes deformations without large-scale rupture and maintains structural stability when exposed to mechanical stress. The theoretical analysis shows that the nuclear envelope is elastic and exhibits the characteristics of a continuous two-dimensional solid. [Rowat, Foster, Nielsen, Weiss, Ipsen, *J. Roy. Soc. Interface* 2, 63-9, 2005; highlighted in *J. Exp. Biol.* 208,vii, 2005].

MEMPHYS's members have in recent yeears contributed to giving an overview of our current understanding of complex membrane structure and function (ø) via a number of review papers [Jacobson, Mouritsen, Anderson, *Nature Cell. Biol.* 9, 7-14, 2006; Khandelia, Ipsen, Mouritsen, *Biochim. Biophys. Acta* 1778, 1528-1536, 2008; Mouritsen, *Biochim. Biophys. Acta* 1798, 1286-1288, 2010; Bagatolli, Ipsen, Simonsen, Mouritsen, *Prog. Lip. Res.* 49, 378-389, 2010; Mouritsen, *Cold Spring Harb. Perspect. Biol.* 3, 33-47, 2011; Mouritsen, *Phys. Chem. Chem. Phys.* 13, 19195-19205, 2011]. In particular lateral membrane structure, lipid domains, and rafts have been considered and it has been pointed out that the concept of rafts is now at a crossroad between physics and cell biology. Membrane lateral heterogeneity is becoming accepted as a requirement for the function of biological membranes, although the notion of rafts still needs to be substantiated. However, the tools are now becoming available to study biological membranes as structured liquids that are partially ordered in space and time. In this picture, the concept of the liquid-ordered phase, originally proposed by workers at MEMPHYS, continues to be a cornerstone.

Technological applications of membranes

As an example of a biomedical application of lipid-bilayer membranes, mention could be made of stealth liposomes that can be used as drug-delivery vehicles, e.g., in cancer therapy (å). This kind of systems is an example of nano-medicine [Mouritsen, *Eur. J. Lipid Sci. Technol.* 113, 1174-1187, 2011]. The escape of encapsulated anticancer drugs from liposomes by passive diffusion often leads to suboptimal drug concentrations in the cancer tissue, therefore calling for effective trigger mechanisms to release the drug at the target. The drug could be an encapsulated entity or an integral part of the liposomal carrier in the form of one or several lipid prodrugs that are turned into active drugs at the target [Arouri, Mouritsen, *J. Liposome Res.* 21, 296-305, 2011]. A promising trigger mechanism is secretory phospholipase A_2, an enzyme that is upregulated in various cancer cells. The optimization of such liposomal drug-delivery systems is critically dependent on an understanding of the physico-chemical properties of the liposomes, the peculiar kinetics of the phospholipase, and the mechanisms of stabilization *vs* destabilization of lipid-bilayer structures.

Outreach activities

Many MEMPHYS members have been engaged in a variety of outreach activities aimed at stimulating young people's interest in science, educating teachers, and informing the public about science in general and about the research at MEMPHYS in particular. Among the many activities, the following can be mentioned: highschool science labs, popular talks, open house arrangements, popu-

lar articles and books, TV and radio programs, science theatre and ballet, national science fairs, Forskningens Døgn, Peoples' University, and Naturvidenskabsfestival. Members of MEMPHYS have taken the initiative to two unusual modes of outreach. One is *natnet.dk* (www.natnet.dk) which is a multi-media internet-based database of so-called `science challenges' for high-school students within physics, chemistry, mathematics, and biology. The other is *Science in Your Eyes* (www.scienceinyoureyes.com), cf. **Fig. 2**, which is an online image gallery showcasing the capabilities of modern imaging techniques, including examples from confocal fluorescence microscopy and atomic force microscopy. The themes are taken from the world of biophysics – cell membranes, fats, proteins and liposomes – with emphasis on the aesthetic nature of cellular and subcellular structure. The site is now an integral part of the Danish national homepage for bioimaging (www.bioimaging.dk).

MEMPHYS statistics

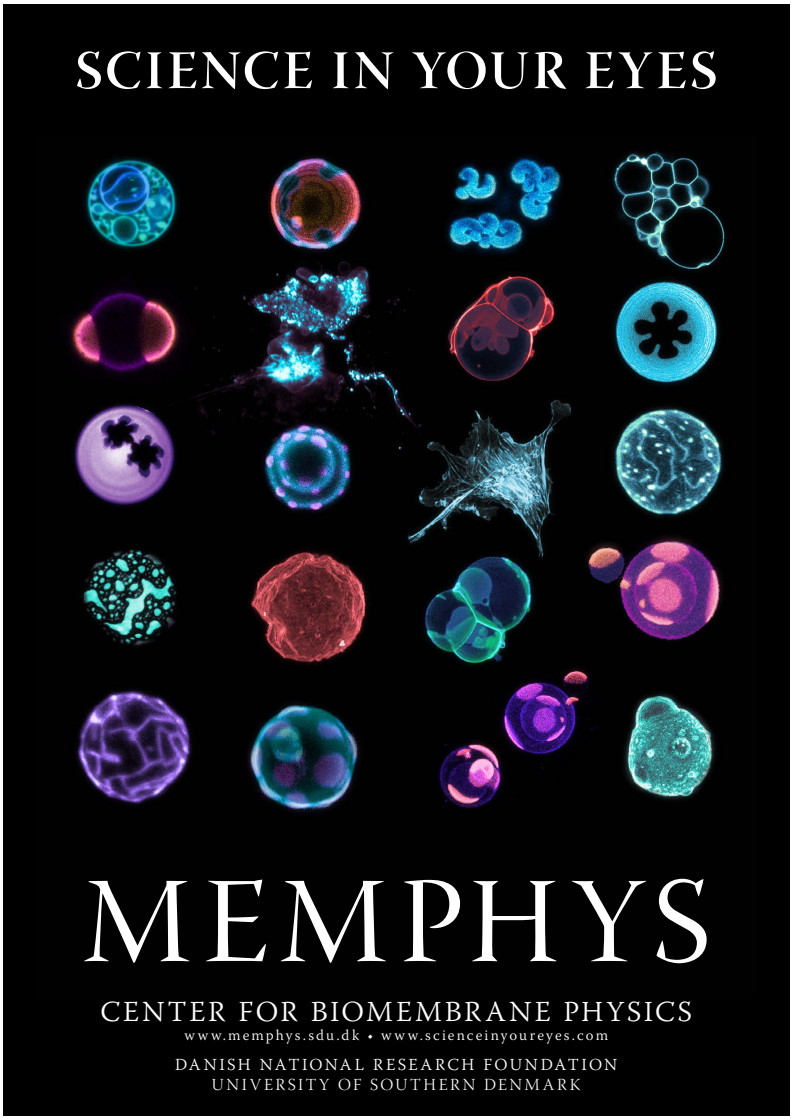
Full accounts of the activities at MEMPHYS in the period 2001-2011 can be found in [1]. Key numbers and statistics are quoted in **Table 4**. A measure of the diversity in the research approaches is reflected in the fact that publications emanating from MEMPHYS over the last ten years have appeared in more than one hundred different types of international scientific journals.

Table 4. Some MEMPHYS key numbers 2001-2011

≥ 400 original articles and review articles (peer review)
≥ 150 published proceedings
≥ 30 book chapters
~ 5 books
≥ 50 popular articles and books
~ 10 cover stories
30-50 seminars/visitors per year
Training of ~ 50 PhD students

The future of MEMPHYS

After the termination of the Center of Excellence Grant from the National Danish Research Foundation, MEMPHYS continues to exist as a center with an infrastructure built around research collaborations and a large common experimental laboratory. Recent developments include a massive built-up of advanced biophotonics instrumentation and surface-sensitive equipment for use in bioimaging. The new experimental facilities are an integral part of the new national core facility, DaMBIC-Danish Center for Molecular Biomedical Imaging (www.dambic.dk), which is based on an initiative taken by MEMPHYS in collaboration with researchers at the Faculty of Medical Sciences at SDU. MEMPHYS is now also part of a national center for nanomedicine, Lundbeckfondens Center of Excellence *NanoCAN* (Nanomedicine Research Center for Cancer Stem Cell Targeting Therapeutics) that is formed as collaboration between researchers at the Faculties of Natural Sciences and Medical Sciences at SDU. MEMPHYS continues to be strongly involved in theoretical modelling and simulation of biomolecular systems and therefore remains connected to the national E-science infrastructure by partnership with the Danish Center for Scientific Computing.



Figur 2. MEMPHYS poster for 2011. Posters from other years can be downloaded from www.scienceinyoureyes.com.

Future research directions of MEMPHYS will be diverse and constantly changing, with a solid basis in fundamental physics and physical chemistry. Recent research interests fall within food and dairy science, gastrophysics, pharmaceutical research involving particulate drug formulations, nano-medicine, biomedical imaging, and non-equilibrium membranes.

Some thoughts regarding Centers of Excellence [2]

To create and run a successful Center of Excellence (CoE) is so much more challenging and exciting than doing a bunch of research projects. A CoE is a creative and stimulating home for open-minded scientists and students who share a common vision and fascination for their science that go way beyond doing the individual research projects and reaching the milestones of the research plan. It kindred spirit should be dynamic, bold, daring, controversial, and unorthodox. The commitment is to contribute both to the frontiers of science, as well as to maintain and renew the scientific undercurrent from which new and ingenious ideas, talents, and new disciplines can emerge and be the nuclei for future CoEs.

Centers should therefore not isolate themselves from their host institutions but integrate, inspire, and nurture their environment. This is an obligation as well as a privilege. In my view, a successful CoE should during its ten years of existence constantly renew itself and at the end of the period have transformed into something quite different from the beginning and repeatedly have changed research directions and possibly also have changed field. A CoE is in this way much more than the science it is doing but also a particular attitude to academia and society.

Invariably, there will be groups at the host institution of a CoE that may feel and even express that the large influx of free funds and activity typically associated with a CoE is unfair and possibly devastating for the vigorous development of other research activities. This is something the CoE has to take seriously up front and deal with it, e.g., by making clear and demonstrating by example that a center is added and synergetic value to the university, both in terms of research, teaching, and an active academic environment. Means to this end can be to engage non-center members via collaborative projects, co-supervision of students, making the center's equipment and laboratories available to others, and always let center activities be open to all interested. Another instrument is organization of scientific workshops that not only feature the work of the center but also cover more broadly and invite scientists from other fields to contribute. Often the inspiration comes from the most unexpected directions. Be inclusive rather than exclusive. The most durable measure is of course to hire members into the center who are not scientific one-trick-ponies but who are academically well versed in several disciplines.

If I should mention a singular crucial element behind the success of MEMPHYS, besides the actual science we did, it is the establishment and running of an experimental laboratory that is truly common for all center members. During the lifetime of the center this laboratory grew to become the possibly largest and best-equipped molecular biophysics laboratory in Denmark. Visiting scientists from across the world are impressed with the fact that we managed to make such a set-up where all center scientists share equipment and have a common responsibility for installing, maintaining, and upgrading cutting-edge instruments. This unique way of running a large laboratory is probably why we were also successful in establishing a core facility for bioimaging jointly with the Medical School at SDU.

References

[1] Activity Reports from MEMPHYS, see http://www.memphys.sdu.dk/progressreports/

[2] Extract of: DG Center of Excellence - a dynamic concept beyond the research project (O. G. Mouritsen) *DG Info* **11**, 2-4 (2011).