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Preface

Dear Master’s Thesis student,

Facing the choice of Master Project can be bewildering and daunting. Maybe, you had already made up your mind for a particular field a long time ago, but have then been tempted by other possibilities, maybe you are simply bewildered by the many possibilities whether it is a more technological or a more patient oriented project, or maybe you have not considered anything at all yet. Take it easy, but be ready!

There is plenty of time to consider where you want to write your thesis and to contact different supervisors to get further information. However, as there is a limit to the number of students on each project, you stand a greater chance of getting the thesis you want, the earlier you get going. You should also consider whether you wish to write your thesis alone or in collaboration with another student (in some instances, this may be a requirement).

This thesis catalogue is intended to provide inspiration and help you to get the thesis that suits you. The catalogue is structured in such a way that each research group, along with its supervisors, is described with examples of projects.

As you will not be writing your thesis until next year, these descriptions should be viewed as tentative only, the reason being that future research results may alter the projects. Therefore, the final projects will be defined by agreement with the students immediately before the projects are to start.

As a master’s thesis student at the Department of Pharmacy, you take part in the daily work along with the other employees and are carrying out a delimited subproject under one of the current research projects. During this process, you will have one or more supervisors who will be highly familiar with your project. We would be looking forward to welcome you at the department for any of our projects spanning from "molecule to society".

Hopefully, we will be able to provide you with a taste of the exciting and mind-blowing field of research. And assist you in the transition from student to researcher. Anyways, we guarantee that you will be challenged on the way, but will also provide a helpful and stimulating work environment to support you.

You are always welcome to contact the Master’s Thesis contact persons, if you require further information.

Best wishes,

Flemming Madsen
Head of Department of Pharmacy
Microscale Analytical Systems

The main research focus of the Kutter group is the development of advanced analytical separation and sample preparation tools to gather (bio)chemical information for the potential benefit of pharmaceutical treatment and medical care.

Our research group provides analytical chemical tools for tackling many challenges in pharmaceutical and medical research, especially in situations where sample size is limited, where the molecules of interest are only present in small concentrations, or where a large number of sample needs to be processed in a short time frame. To enable this cutting-edge analysis we develop and apply technologies such as micro-nano engineering, microfluidics, and polymer science for next generation high-throughput workflows, e.g., in protein characterization, peptide analysis, drug transport studies, and drug delivery.

“We are experts in qualitative and (validated) quantitative analytical chemistry. We are developing new ways to perform chemical analysis, while pushing traditional approaches to their limits.” says professor and group leader Jörg P. Kutter.

Main focal areas of the group are modern separation techniques (e.g. LC, CE) coupled to mass spectrometry, advanced sample preparation methods (e.g., μSPE, μEME), traditional spectroscopic techniques (e.g., NMR, IR, Raman) and a range of microfluidics-driven techniques (e.g., droplets, micro-nano particle fabrication); furthermore, we apply ex vivo models and develop microphysiological in vitro models (i.e., organ-on-a-chip) that are all intended to understand drug-target interactions, investigate drug metabolism, and getting insight into disease progression and therapeutic efficacies.

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Project examples
Cerebral stroke model on-a-chip
In the human body a sudden loss of oxygen supply can cause irreversible impairment like ischemic brain damage. The brain requires a high amount of energy, which it derives from oxygen and glucose, and is supplied by cerebral blood. Since the brain is not capable to reserve energy it is highly susceptible to interruption of blood flow. A blockade of the cerebral blood flow is called stroke and immediately causes brain damage.

The blood-brain-barrier (BBB) separates the cerebral blood from the brain and constitutes the physical and metabolic barrier, while in the early stage of ischemic stroke it loses its integrity leading to infiltration of local inflammatory cells and post-ischemic oedema and swelling. In recent years, microfluidic devices have been proven to be an ideal platform to mimic ‘in vivo-like’ conditions for cell cultures. Such micro-devices enable precise temporal and spatial control, which allows the complex formation of biophysical and biochemical microenvironments for in vitro studies.

The goal of the thesis project is to establish an in-vitro model of the BBB endothelial cells on a micro-scaled device. With a novel approach developed in our research group, hypoxic conditions will be recreated to enable the study of cerebral ischemia and find potential drug candidates for treatment.

Supervisors: Drago Sticker, Jörg Kutter, Birger Brodin
No of students: 1-2

Bridging the gap between preclinical models and clinical studies in drug discovery
The field of drug discovery and development is facing a major challenge of rising costs and declining efficiency of drug research and development. Drug failures are primarily due to the poor preclinical predictive power of existing models.

In vitro models lack the complexity of a living multicellular organism and fail to mimic the native environment, leading to simplistic models of limited value for drug discovery. The major concern is the dedifferentiation of primary cells, resulting in misleading experimental results. To overcome this limitation, microphysiological systems have recently been introduced, which aim to mimic the physiology and structure on the organ level (so called organ-on-a-chip). These systems enable the creation of complex in vitro models by co-cultivation of different types of cells; provide biochemical and mechanical cues by e.g. circulating medium flow and therefore preserve cellular functionality. The blood-brain barrier (BBB) is a prominent example where all these factors are of crucial importance. Microphysiological systems are ideally suited to mimic the BBB environment by applying constant medium flow (shear force), incorporate multiple cell types and brain specific extracellular matrix. Organ-on-a-chip technology significantly changed the drug discovery paradigm and
could in future decrease the translational failure in drug development.

**Supervisors:** Drago Sticker, Hans Christian Cederberg Helms, Jörg Kutter, Birger Brodin
**Possibility to conduct parts of the project abroad (Austria or Netherlands).**

**No of students:** 1

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**Cannabinoid transport across the blood-brain-barrier**

Cannabinoid-derived pharmaceuticals are widely used to treat disease or alleviate symptoms, but their transport mechanism across the blood-brain-barrier is rather unexplored. For an effective treatment of neurological diseases the compounds need to reach the central nervous system. During this master project the role of co-factors (terpenophenolic compounds known as phytocannabinoids) in the permeability of the cannabinoid across the BBB will be investigated. Transport studies will be conducted on whole plant extracts and compared to pharmaceutical grade cannabinoids (e.g. THC, CBD).

**Supervisors:** Drago Sticker, Hans Christian Cederberg Helms, Jörg Kutter, Birger Brodin, Nickolaj J. Petersen

**Industrial collaboration with Canna therapeutic ApS.**

**No of students:** 1

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**Protein biochips for in-vitro characterization of biochemical activities**

The ability to immobilize biomolecules at specific locations on the surface of solid supports is essential in many biochip applications. Biochips featuring immobilized biomolecules can be used to identify binding proteins from biological samples or as enzymatic micro-reactors for the screening of drugs and their metabolites. Thiol-ene has a very high potential as a biochip substrate for the in-vitro characterization of biochemical activities. The thiol-ene reaction can be used to form cross-linked polymer substrates for microfluidic fabrication as well as to covalently attach recognition molecules at the surface of these substrates. We have recently demonstrated that the optical properties of thiol-ene polymers combined with the ease of modifying their surface for the site specific attachment of recognition molecules make them ideal candidates in many biochip applications. Since the reaction is photo-initiated, specific patterns can be obtained by exposing only the desired areas using photomasks as shown in the Figure. The goal of this project is to use thiol-ene biochips to create enzymatic reactors for the screening of drugs and their metabolites. Enzymes can be immobilized rapidly and specifically via their non-essential thiols to thiol-ene substrates featuring an excess of allyl groups. Such enzyme micro-reactors combined with fluorescence or electrochemical detection provide an attractive method of generating and detecting drug metabolites.

**Supervisors:** Jörg P. Kutter and Drago Sticker

**No of students:** 1-2
Micro-reactors for the rapid screening of endocrine disruptors

In human phase I drug metabolism, enzymes catalyse the oxidation of xenobiotics to facilitate their excretion or further metabolism. The study of these metabolic pathways is highly relevant for toxicity assessment and drug discovery. Enzyme micro-reactors based on chip technology are especially attractive for this type of studies because they can provide continuous generation of enzyme products and allow for the re-use of immobilized enzymes. In this project, human cytochrome P450 (CYP) recombinant enzymes will be immobilized on thiol-ene monoliths through covalent and non-covalent interactions. Several strategies will be employed to enhance immobilized CYP activity, such as the integration of gold, gold-sputtered electrodes in the polymer matrix as a source of electrons for the enzymatic reactions and the use of additives, such as nanoparticles, in the monolith to enhance the conductivity and stability of the immobilized CYPs. The activity of the immobilized enzymes will be thoroughly investigated under various conditions. The immobilized enzymatic micro-reactors will be used for the in-vitro screening ofazole antifungal drugs.

Supervisors: Jörg P. Kutter, Drago Sticker and Andreas Kreitschmann
No of students: 1

Lab-on-a-Chip for next-generation sample separation and analysis

The analysis of complex protein mixtures is of tremendous importance for pharmaceutical drug discovery. To identify potential drug targets, complex mixtures such as blood, cell cytoplasm or secretome have to be screened. Such high-throughput screenings involve large quantities of samples and compounds, which are only available in small amounts. Therefore the sample preparation has to be miniaturized, integrated and automated. Ideal platforms to meet these requirements are microfluidic devices, which allow the precise control of chemical reactions and the integration of other functional elements (so-called lab-on-a-chip). For instance, microchip liquid chromatography provides high separation efficiency, resolution, and sensitivity. During the project a microscaled device capable of chromatographic separation will be further developed to enable direct coupling to highly sensitive analytical methods such as tandem mass spectrometry (MS/MS).

Possibility to conduct parts of the project abroad (Finland).
Supervisors: Drago Sticker, Jörg Kutter, Nickolaj J. Petersen
No of students: 1
Microchip-based chiral separation of racemic drugs
Have you heard of Thalidomide? Thalidomide was sold as a mild sedative, especially targeted to pregnant women and, for the most part, worked as advertised. What they didn’t consider was the fact that the drug was produced as a racemic mixture (i.e. a mixture of two stereoisomers), where one of the stereoforms had no pharmacological effect at all, but gave severe adverse effect in the form of foetal deformations. To make sure that this does not happen again drug companies take great care to screen and separate/isolate their drugs based on chirality (“handedness”). The goal of this project will be to develop and produce a microfluidic chip for chiral separation of racemic mixtures of drugs and/or their metabolites. The separation will be chip-based capillary electrophoresis and the student(s) will be allowed freedom in designing both the actual microfluidic device and the mode of separation. Depending on the early success of the project, collaboration with Andreas Kretschmann from the Toxicology group will give access to racemic azole antifungal drugs and their metabolites, where chiral screening is of outmost importance.

Supervisors: Jörg Kutter, Drago Sticker, Andreas Kretschmann
No of students: 1

DRUG-DISCOVERY-μCHIP. Chemical stability, membrane permeability, protein binding, and in-vitro metabolism of new drug substances measured in a single device
For the development of new drug substances, investigations into their chemical stability, membrane permeability, protein binding, and in-vitro metabolism are mandatory. Traditionally, all these investigations are conducted in separate assays, and this represents a major challenge for the pharmaceutical industry in terms of cost, efficiency, time, and sustainability. In the current project, a micro-chip system is to be developed, which can be used for all the above mentioned measurements, and which can be extended for even more sophisticated experiments in the future. This micro-chip system, termed DRUG-DISCOVERY-μCHIP (DDμC), will consist of (a) an artificial liquid membrane unit, (b) an electrophoresis channel unit, and (c) a mass spectrometry interface unit. The purpose of the artificial liquid membrane unit is to perform membrane permeability assays, and to exclude biological material from entering the electrophoresis channel unit during protein binding and in-vitro metabolism assays. The purpose of the electrophoresis channel unit is to separate drug substances and their metabolites prior to the final detection by mass spectrometry, and the purpose of the mass spectrometry interface unit is to couple the DDμC to the mass spectrometer. This project involves cutting-edge technical development, based on our earlier research experiences, optimization, and application to selected existing drug substances for initial testing of data reliability.

Supervisors: Nickolaj J. Petersen, Drago Sticker, Jörg Kutter, and Stig Pedersen-Bjerregaard
No of students: 1-2
Quality assurance and development of Plant medicine - Cannabis

Focus will be on characterizing cannabis based drug raw materials and products and developing possible new compounds from medical and industrial hemp. Cannabis based plant medicines are rapidly expanding all over the world not the least due to the increased legalization taking place. Due to collaboration with industry (Canna Therapeutic) and the two pharmaceutical departments at the Faculty of Health and Medicine at the University of Copenhagen it is possible to perform studies on qualitative and quantitative profiling of Cannabis species. The project will continue earlier work on cinchona (quinine alkaloids) and Cannabis, where validated homogenization, extraction, and analytical methods have been developed and implemented. We will define the final project with the company, when the time comes. Part or the project could be isolating interesting peaks and determine their structure, and if possible their biological activity.

Methods used: HPLC-UV, HPLC-MS, GC-MS and NMR.

Supervisor: Nickolaj Petersen and Claus Cornett in collaboration with Canna therapeutic.

No of students: 1-2

Grasshopper project 1. Grasshopper model for drug permeation and metabolism studies in brain

Small animal models are a tool in early drug discovery to predict drug uptake and metabolism. Recently, the grasshopper Schistocerca gregaria has been introduced as a model to investigate drug uptake and metabolism in the grasshopper brain. To estimate drug and drug metabolite concentrations in the grasshopper brain it is crucial to assess the size of the brain. In humans, one of the most abundant compounds in brain is the amino acid n-acetyl aspartate (NAA), which we also found on the grasshopper brain. The aim of the project is to develop and validate an LC-MS method to quantify NAA in the grasshopper brain. Therefore, you will be trained in handling the grasshopper, dissecting the grasshopper brain and preparing biological samples from the brain matrix. After validating your quantitative method and quantifying NAA in the grasshopper brain, you will be able to compare your analytical approach with other assessment strategies to estimate the size of the grasshopper brain which we have been investigating in our lab, such as measuring the brain size with microscopy or quantifying the protein concentration of the brain.
Grasshopper project 2. Upgrading the grasshopper in vivo model
Finding human drug metabolizing enzymes in other species, such as insects, can reduce the number of animal experiments in pre-clinical trials in drug discovery and development. Within this project, you will work with the grasshopper Schistocerca gregaria, to find relevant drug metabolizing enzymes in the grasshopper’s “blood” – the hemolymph. You will learn, how to inject drug compounds into the grasshopper and how to collect samples, and how to set up and run enzyme assays, and how to prepare these samples for instrumental analysis with LC-MS or UV detection. Finally, you will be able to evaluate drug metabolism in the grasshopper’s hemolymph and conclude on the relevance of your findings for the grasshopper model in early drug discovery and development.

Supervisors: Nickolaj Petersen and Claus Cornett
No of students: 1-2

Grasshopper project 3. Upgrading the grasshopper in vitro model
The most important group of enzymes relevant for drug metabolism in humans are CYP P450 enzymes. These enzymes can be extracted from tissue as microsomes and thus, can be used as in vitro test system to study enzyme activity and drug metabolism in high throughput. Recently, a functional homologue of the human microsomal CYP 3A4 has been found in in vivo experiments in the grasshopper. The aim of the project is to develop a functioning microsomal preparation from relevant grasshopper organs. You will learn to dissect grasshopper organs and to prepare microsomes. You will be trained in characterizing the quality of your microsomal preparation by assaying the protein concentration and the overall CYP P450 activity. Finally, you will evaluate the performance of your microsomal preparation by investigating the metabolism of a drug compound. You will be able to compare the results of your experiments with human liver microsomes and rat liver microsomes.

Supervisors: Nickolaj Petersen, Claus Cornett and Andreas Kretschmann of students: 1-2
Capillary electrophoresis - electrospray ionization mass spectrometry, CE-ESI-MS
Recently we developed a new and robust CE-ESI-MS interface. The interface is "plug and play" and with basically no dead-volume at the electric contact that is supplied only 6 mm from the ESI spray tip. This project involves testing the new device for the fast quantitation of biogenic amines in red wine (see website).
Supervisor: Nickolaj J. Petersen
No of students: 1

Online sample preparation coupled to electrospray ionization – mass spectrometry
We have recently demonstrated a new way of performing continuous liquid-liquid extraction on microfluidic devices. Target analytes from biological sample can be selectively extracted from a flowing sample solution, through a 25 μm thick supported liquid membrane (SLM) and into a flowing acceptor solution. The extraction across the organic SLM is controlled by an applied electric potential across the membrane (electro membrane extraction). The flowing acceptor solution can be coupled online to a mass spectrometer for continuous monitoring. In this project you will use the devices for studying the metabolism of drugs in real time.

The online coupling of metabolism with MS is particularly interesting (see website), because it may reveal short-lived reactive metabolites as well as the kinetics of the metabolism.
Supervisor: Nickolaj J. Petersen
No of students: 1

2D LC-MS in complex formulations and bioanalysis.
Quantification and identifications of impurities is tricky in complex matrices due to interferences. This problem is intensified in formulations with low dose, potent drug compounds due to the reporting limits for impurities given in the ICH guidelines.
By using two-dimensional liquid chromatography (2D LC) the selectivity between analytes and matrix may be enhanced giving fewer problems with interferences. A master thesis project could consist of an evaluation of the usability of 2D LC for: 1) Quantification of low level impurities in formulations with 2D LC-UV, 2) Elucidation of impurity identity by 2D LC-MS.
The project will be conducted in Analytical Support at LEO Pharma in Ballerup, and the content may be adjusted based on on-going activities.
Analytical Support at LEO Pharma is a part of Pharmaceutical Technologies in the LEO Pharma R&D organization. We have strong competencies within separation techniques (UHPLC, GC, and SFC), solid state NMR and MS. We provide analytical support to R&D enabling LEO Pharma to deliver new innovative products and solutions.
Internal supervisors: Nickolaj J. Petersen
Protein Analysis Group

Homepage: http://pharmacy.ku.dk/research/section_analytical_biosciences/protein_analysis-lab/

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Biopharmaceuticals (i.e. protein-based drugs) account for the most rapidly growing type of drugs in the pharmaceutical industry. Through the thesis projects below, you will get hands-on laboratory experience of how to work and handle proteins and learn state-of-the-art techniques for analysis and characterization of biopharmaceuticals (protein chemistry, biophysics, liquid chromatography and mass spectrometry).

Structural analysis of biopharmaceuticals

Analysis of the primary (covalent) and higher-order structure (conformation) of protein-based drugs is critical for both understanding molecular action as well as monitoring and comparing drug product quality. In this suite of projects you will apply a new powerful analytical technology that uses mass spectrometry to measure the hydrogen/deuterium exchange of proteins in solution (HDX-MS) and is thus capable of producing detailed information on the structure of biopharmaceuticals. You will use this technique to analyze and compare the structural properties of new potential protein drugs in development and thus help provide a critical molecular understanding of their pharmaceutical properties and function in the human body. Work will be done in close collaboration with researchers at UCPH or industrial collaborators such as Biogen Idec (USA), Roche (Germany) and Novo Nordisk.
(Denmark), depending on the project. There is typically also the possibility to perform parts of the project at the industrial collaborator. 

**Supervisors:** Kasper Rand and Tam Nguyen.  
**No. of students:** 1-2

**Understanding the binding of biopharmaceuticals to target protein receptors**  
We have recently developed a method whereby the binding of small molecule ligands or large protein based drugs to naturally occurring protein receptors can be studied in great structural detail. By using mass spectrometry, we measure the hydrogen/deuterium exchange (HDX-MS) of the protein receptor in the absence and presence of a single or a panel of potential ligands. We can thus map the binding site of ligands on the protein receptor and study the structural effects of binding. In this project, you will use this method to study both the binding of new small molecule drugs and large protein-based drugs to an important new pharmacologically interesting protein receptors. Work will be conducted in collaboration with academic or industry collaborators.  
**Supervisor:** Kasper D. Rand  
**No of students:** 1-2

**Quantitative analysis of biopharmaceuticals in biological samples**  
Quantification of protein drugs in serum currently relies on biochemical assays which require extensive optimization and sample preparation. A new and better technique for protein quantitation in human blood could be a great asset to improve stability and pharmacokinetic studies during biopharmaceutical drug development. In this context, the huge potential of liquid chromatography (LC) or capillary-electrophoresis coupled with tandem MS to give sensitive, specific and accurate quantitative measurements of biopharmaceuticals in serum have not been harnessed. This project will focus on developing a novel LC-MS / CE-MS strategy for quantification of therapeutic peptides and proteins in serum, possibly in collaboration with industry.  
**Supervisors:** Kasper Rand and Tam Nguyen  
**No of students:** 1

**Coupling microfluidics and mass spectrometry for improved analysis of proteins**  
Analysis of protein drugs requires specialized sample handling and advanced analytical chemistry techniques including liquid chromatography and mass spectrometry. Furthermore, proteins often contain covalent modifications (e.g. glycosylations, disulfide bonds) which render them difficult to analyze. In this project we will explore the use of miniaturized analytical platforms, or microfluidic chips, to perform rapid and automated preparation of protein samples (sample concentration, enzymatic reactions, chromatographic separation) for analysis by mass spectrometry. You will learn how to implement protein chemistry and liquid chromatography on a microfluidic device coupled to state-of-the-art protein analysis by mass spectrometry. The project will be done in collaboration with Jorg Kutter of the Microscale Analytical Systems group.  
**Supervisors:** Kasper D. Rand.  
**No of students:** 1-2.
Examples of publications (links to websites) resulting from past master thesis projects in the group

Trabjerg et al. 2017
Qiu et al. 2016
Mistarz et al. 2016
Trabjerg et al. 2015
Hongjian et al. 2014
Toxicology and Drug Metabolism Group

We perform experimental research revealing the occurrence, fate, and toxicological and endocrine effects of xenobiotics, especially pharmaceuticals, with the aim of assessing their risks to humans and biota. We apply various in vitro, ex vivo and in vivo assays and advanced analytical techniques, such as LC-MS and mass spectrometry imaging.

See website for Toxicology and Drug Metabolism Laboratory

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Please find examples of projects in Toxicology and Drug Metabolism Laboratory on the following pages.
Project examples

Endocrine disrupting pharmaceuticals

Public awareness concerning endocrine disrupting drugs has increased the last few years, as these are presumed to affect reproductive ability and to increase the occurrence of hormone-dependent cancers. Endocrine disrupting drugs are, however, generally much less potent than physiological hormones, and therefore there is doubt as to whether or not these substances can affect humans. Recent research indicate that simultaneous exposure to a cocktail of endocrine disrupting drugs, each at a concentration below the effect level, might result in a significant additive or synergistic effect. The present project wishes to uncover interactions between selected hormone-disturbing substances in various in vitro test systems and/or animal experiments with selected endpoints, including hormonal activity, changes in aromatase activity and the impacts of endocrine disrupting drugs on endogenous hormone metabolism.

**Supervisor: Bjarne Stylishave, Cecille Hurup Munkbøl.** No of students: 1-2

Development of a microfluidic assay for testing endocrine disrupting potential of pharmaceuticals

Certain groups of drugs, such as azole fungicides, have enormous potential to disrupt the hormone system in humans and are suspected to contribute to the worldwide increasing incidence of birth defects, infertility, cancer and obesity. However, information on unintended effects of drugs on the human hormone system is in general scarce.

One problem is that existing in vitro assays, which can measure the endocrine disrupting potency of drugs, are too time demanding and too expensive. The goal of this Master thesis is therefore the development of a microfluidic assay for the high throughput screening of drugs for their endocrine disrupting properties. Within this project an OECD standardized cell-based in vitro assay (H295R steroidogenesis assay), which can detect effects of drugs on the steroid hormone system, will be miniaturized on a microchip. The microassay will be validated with azole antifungal drugs, which are known to inhibit the synthesis of hormones like the sex steroids testosterone and estradiol.

**Supervisors: Cecille Hurup Munkbøl, Andreas Kretschmann, Drago Sticker, Joerg P. Kutter.** No. of students: 1-2
Endocrine toxicity of drug enantiomers and metabolites

Increasing evidence exists that certain pharmaceuticals disturb the human hormone system and may contribute to endocrine related diseases like infertility, obesity and different types of cancers. It is well known that enantiomers of drugs can differ strongly in their biological activity. Furthermore, drug metabolites formed in the body can be active and possess higher activity than the parent compound. Very little is known about how the endocrine disrupting potency of a drug depends on its enantiomeric form and its metabolic products.

The focus of this project are chiral drugs like azole fungicides, which are used for the treatment of severe systemic and superficial fungal infections. The goal of this project is to elucidate the endocrine toxicity of azole enantiomers and metabolites through pharmacokinetic and dynamic studies. The practical work includes enzyme and cell based in vitro assays as well as in vivo experiments. A large part of the project is the development of analytical chemical methods for the identification and quantification of enantiomers and metabolites in different biological matrices with enantioselective HPLC-MS/MS.

Supervisors: Andreas Kretschmann, Cecilie Hurup Hansen, Bjarne Styrisheve, Claus Cornett. No. of students: 1-2

Azole transport in the human term placenta

Azole drugs are highly efficient drugs against fungal infections. However, azoles are notoriously known for their endocrine effects. Since pregnant women are at increased risk of experiencing fungal infections, there is a growing concern that azoles will pass the placenta and exert endocrine effects on the developing fetus. Furthermore, the azoles may obstruct the balance between androgens and estrogens, tightly regulated by the placenta. In this project, we investigate the placental transport of azole fungicides widely used during pregnancy, and the ability of the placenta to regulate sex steroid balance during treatment to azole antifungal drugs.

Supervisors: Bjarne Styrisheve, Andreas Kretschmann. No of students: 1-2
Pancreatic infections: Local and systemic antibiotics in infected walled-off pancreatic necrosis.
Severe acute pancreatitis is characterized by organ dysfunction and necrosis of the pancreas. Mortality is high, up to 20%. Infection in the pancreas is a serious complication and is associated with high mortality, up to 20%. After 3-4 weeks the necrosis becomes encapsulated a so-called walled-off necrosis (WON). The primary treatment is systemic antibiotics. However, little is known about the penetration of antibiotics into the necrosis and the ability of antibiotics to stop the infection. Several factors may influence the activity of the antibiotics, such as the fibrous capsule surrounding the necrosis, the size and the physic-chemical milieu of the necrosis. Using 2D-DESI-imaging and LCMS/MS, this project aims to investigate the distribution of the most commonly used antibiotics in pancreatic necrosis. The purpose is to identify the most effective treatment, thereby decreasing mortality and morbidity in patients suffering from pancreatic necrosis.


Danida project: Green Resource Innovations For Livelihood Improvement (GRILI)
Medicinal plants as green resource products (GRPs) are used as traditional medicine in Tanzania, and are key components for primary health care and livelihoods of more than 50% of the inhabitants. Unfortunately, many herbal products are adulterated with drugs, such as antibiotics, antimalarials and mild analgesics. The present study aims to investigate the adulteration of herbal drugs from Tanzania, by analysing the presence of different drug classes in herbal medicines collected from markets and health providers in Tanzania. Samples will be collected and extracted in Tanzania and transported to Denmark, where the samples will be analysed. The project included approximately 3 months of field work in Tanzania.

Mass spectrometry imaging of drugs in tissue sections from mice
Using a new technique with mass spectrometry, Desorption Electrospray Ionization Imaging (DESI-MS), it is possible to image the distribution of compounds on a surface. In this way, we can follow a drug dosed to a mouse, creating images of the mouse that show where in the body the drug goes, and how it is metabolized. In this project, a mouse will be dosed with a drug, and subsequently the whole mouse or single organs (brain, liver and kidney) will be cut in thin slices which are then analysed with DESI-MS. The aim of the project is to image where the drug goes and in which doses we can see the drug and its metabolites. The project will be planned in collaboration with pharmacologists or medicinal chemists.

Supervisor: Christian Janfelt. No. of students: 2

Mass spectrometry imaging of natural compounds in cannabis
The use of medical cannabis is increasing worldwide, and recently, the Danish government has sanctioned a 4-year pilot programme for the prescription of medical cannabis to Danish patients. More than 500 natural compounds have been isolated from Cannabis Sativa, including the medically important cannabinoids, terpenes and flavonoids. For a deeper understanding of this complex composition, and to ensure products of high and consistent quality, better analytical tools are needed. The aim of this project is to apply mass spectrometry imaging to characterize the distribution of active compounds in Cannabis Sativa leaves, to study the compound synthesis in the plant during flowering and the impact of environment and growth conditions on compounds of interest in the plant.

Supervisor: Christian Janfelt. No of students: 2

High-resolution MALDI mass spectrometry imaging for studies in drug delivery
A new laser-based mass spectrometry imaging setup has been installed, which enables mass spectrometry imaging at the cellular level. The technique will be used to study the delivery of drugs across biological membranes and barriers in ex-vivo and possibly in-vivo animal experiments. High resolution mass spectrometry images showing the distributions of drugs and other exogenous compounds relative to endogenous compounds such as membrane lipids will provide information about the efficacy and mechanism of new drug formulations. The project will be planned in collaboration with one of the Drug Delivery groups at the Department of Pharmacy.

Supervisor: Christian Janfelt. No of students: 2
Mass spectrometry imaging in development of skin cancer treatment

The capabilities of DESI and MALDI MSI to image the distribution of endogenous compounds together with a drug and its metabolites make them ideal in the study of skin penetration of drugs. This project takes place in collaboration with Department of Dermatology, Bispebjerg Hospital, which develops new treatments for skin cancer. One approach is to ablate channels in the skin in order to be able to perform local, topical chemotherapy of skin cancer. In this project MALDI imaging and LC-MS will be used to optimize the delivery of one or more chemotherapy agents by imaging of tissue sections from difference depths in the treated skin.

Supervisor: Christian Janfelt. No of students: 2
CNS Drug delivery and Barrier Modelling

We investigate how the absorption, distribution, metabolism and excretion of drugs are influenced by the properties of the barrier tissues in the body. This major focus at present is on transport of compounds through the blood-brain barrier as well as intestinal absorption of drugs and prodrugs via nutrient transporters.

The practical work includes physicochemical studies of drugs, prodrugs and model drugs, ADME studies in cell models and characterization of relevant membrane transport proteins.

The group consists of one associate professor, one assistant professor, one part time research scientist, one postdoc, three PhD students and one technician. We work in a cross-disciplinary fashion, have an international network of collaborators and can offer an exciting work environment.

See website for Drug Transporters in ADME Group

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Project examples

Cell models for screening of CNS drug compounds
Industrial screening of CNS drug compound candidates involves the use of cell culture models. The project concerns characterization of drug transport in cell culture models developed by the group, and investigation of their use in predicting blood-brain barrier permeability. The overall aim of the project is to generate predictive tools for CNS drug development.

Supervisors: Birger Brodin and Lasse Saaby
Max no of students: 3

Drug transport to the brain and the role of the blood-brain barrier
The blood-brain barrier limits uptake of drugs to the brain, and presents a major obstacle in CNS drug development. We investigate transporter expression, receptor-mediated uptake of drug compounds and regulation of barrier properties in the brain endothelium. The overall aim of the project is to investigate how drugs can be delivered to the brain. The masters projects within the area with characterisation of drug, prodrug or model-drug transport across blood-brain barrier model cell lines, as well as characterization of the biological and biophysical mechanisms influencing CNS drug delivery

Supervisor: Birger Brodin
Max no of students: 3

External Master’s projects
Some collaboration with industry may be possible. Previous projects have been performed at Novo Nordisk and Lundbeck.

Earlier Master’s projects: Master’s theses are available upon request.

Drug delivery across the blood-brain barrier using shuttle peptides
A number of peptide drugs for the treatment of brain diseases are available. However, to reach their target site of action, they must pass the blood-brain barrier (BBB). The capillary endothelium comprises the major physical barrier of the BBB and allows only passive permeation of molecules <400 Da. Brain delivery of the larger biopharmaceuticals, which today includes an increasing number of novel peptide-drug entities, is therefore restricted; both due to their large molecular size and hydrophilic nature. Thus, the development of peptide-drugs for the treatment of brain specific diseases requires a delivery strategy for overcoming the endothelial BBB in order to reach its final target within the brain.

Supervisor: Mie Kristensen

Stability of cell-penetrating peptide conjugated therapeutic peptides in biological matrices and their adsorption to plasma proteins
The cell-penetrating peptides (CPPs) comprise a promising tool to facilitate delivery of macromolecular drug entities not only into cells but also across biological barriers, such as the BBB. However, due to their peptide nature they are prone to enzymatic degradation. In addition, cationic CPPs have been demonstrated to adsorb to plasma proteins, thus potentially hindering their cellular uptake.
With the present project, the CPP stability in relevant matrices (physiological buffer, cell culture media, plasma) as well as during incubation with brain endothelial cells (e.g. bEND3 cell line and a primary blood-brain barrier model) will be evaluated using e.g. HPLC, LC-MS, SDS-PAGE, and thin layer chromatography. In addition, CPP interaction with plasma proteins and its effect on uptake into brain endothelial cells will be evaluated via e.g. cell uptake studies and confocal microscopy.

**Supervisor: Mie Kristensen**

**Glycocalyx characterization on in vitro blood-brain barrier models**

The cell-penetrating peptides (CPPs) comprise a promising tool to facilitate delivery of macromolecular drug entities not only into cells but also across biological barriers, such as the BBB. The mechanism by which CPPs translocate across cell membranes is widely discussed and both direct membrane translocation and endocytic uptake has been demonstrated. In addition, a number of studies suggest importance of CPP interactions with cell surface glycosaminoglycans (GAGs) prior endocytic uptake. GAGs are polysaccharide chains attached to a core protein unit making up the proteoglycans within the cell surface glycocalyx.

In order to obtain detailed knowledge on the potential involvement of GAGs for CPP membrane translocation, the glycocalyx layer on the cell culture model used for mechanistic studies must be well characterized. Furthermore, some studies question the presence of glycocalyx in endothelial cell culture.

With the present study we will characterize the glycocalyx surface lining primary mouse brain endothelial cells cultured as monoculture on permeable filters or in co-culture with mouse astrocytes. Employing antibodies and lectins we will identify the GAG composition and protein anchors as well as sugar moieties using confocal microscopy and Eastern blotting. In addition, electron microscopy may be applied to visualize the Glycocalyx morphology.

**Supervisor: Mie Kristensen**

**Max no of students: 3**
Drug Delivery and Biophysics of Biphamaceuticals Group

Our research focus is on the design and development of optimal drug formulations and drug delivery systems, with a specific focus on the delivery of therapeutic peptides and proteins. Our research spans the whole breadth of physicochemical analysis of peptide and protein stability, via design of advanced delivery systems, to the in vivo fate of these delivery systems.

The aim of our research is to elucidate detailed, fundamental, and mechanistic understandings of the challenges associated with the design of drug formulations to obtain optimal and targeted delivery of these drugs, both for injectable and non-injectable routes of administration (e.g. via the gastro-intestinal and respiratory tracts, and via the skin).

More specifically, the research in our group can be described by the following sub-areas:

- Biophysics of peptide and protein formulation, with a special focus on aggregation processes
- Peptide and protein biomaterials
- Oral and pulmonary delivery of peptides and proteins
- Advanced delivery systems, e.g. based on polymers or cell-penetrating peptides
- In vivo assessment, and imaging utilizing radiopharmaceuticals

The group currently consists of one full-time Professor, one joint Professor with the University of British Columbia, two Associate Professors, one Assistant Professor, three Post Docs, one Research Assistant, a scholar student and five PhD students. Typically, more than ten Master students a year join our group.

As a Master student, you will be a part of the group research activities. You can be involved in projects within the group, in industry, or with academic collaboration partners abroad. We have a multitude of opportunities to collaborate and design suitable Master projects – also for extended project periods.

Some generic examples of projects are shown on the following pages. Come talk to us for more details.
Protein and Peptide Self-Assembly: Structures and Mechanisms

Vito Foderà,
Associate Professor, PhD
Email: vito.fodera@sund.ku.dk
If you want to look at my research focus, my network, team and publications, please visit: https://www.vitofodera.com/

Please find below examples of projects within the 4 main areas we are working in.

**NB:** They are only examples, we can design the project that suits you best. Come and talk to us!

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**Role of Protein-Protein interactions in the formation of protein superstructures**

Protein-protein interactions are regulated by the physicochemical properties of the solution. Co-solvents (e.g. alcohols), pH and mechanical stress strongly affect such interactions having as a final result the modification of the aggregation reaction. With this project we want to investigate what is the effect of different parameters on both the kinetics of formation and the structure of superstructures.

**Amyloidogenic protein interaction with cell membranes**

The formation of amyloid fibrils is considered to play a key role in the development of pathologies such as Parkinson’s and Alzheimer’s diseases. New view supports the concept that the interactions of amyloidogenic proteins with cell membranes are a key factor in regulating related toxicity mechanisms. Aim of this project is to directly observe the progression of amyloid fibril formation in the presence of membranes both in synthetic model systems and in living cells.

**Protein-based biomaterials for drug delivery**

A new frontier in protein self-assembly is represented by the analysis of the protein aggregate in terms of its mechanical/structural/thermodynamical properties. This is pivotal for the use of protein aggregates as biomaterials for drug delivery. Aim of this project is to design and produce protein-based materials using different processing methodologies, from electrospinning techniques and bulk methods to microfluidic chips.
Protein stability in pharmaceutical formulations

The presence of protein aggregates in protein drug products is a major concern in pharmaceutical industry. These particles may indeed alter the efficacy of the product. As a consequence, it is of great relevance to isolate and characterize each of these types of particles and evaluate their risk profile. Aim of this project is to produce and analyze homogeneous populations of different protein aggregates originated from insulin formulations.

Therapeutic Peptide and Protein Instability and Formulation
Supervisor: Marco van de Weert  
Marco.vandeweert@sund.ku.dk

Research focus
- Formulation of peptides and proteins
- Physical instability of peptides and proteins
- Peptide and protein aggregation
- Analytical methodology to characterize peptides and proteins
- Immunogenicity of protein therapeutics

Possible projects

Using ligand binding to stabilize proteins against aggregation and adsorption
Ligand binding is a possibly underutilized approach to stabilize proteins against aggregation and adsorption. Can you show it is a viable formulation approach? The project can focus on the instability pathways (biophysics) or on the formulation aspects (excipients and stress factors). Internal project

Characterizing protein-ligand interactions
Ligands may be great stabilizers, but we do need to characterize their binding to the protein - how many ligands bind, how strong, and how does it affect the protein? Many methods are available, but how good are they really? For those interested in proper analytical science, proteins, and a bit of math in the mixture, this is your project! Internal project, but the adventurous student may also spend some time in Rome, Italy

Peptide aggregation
Peptides are similar and different to proteins. Both tend to aggregate. We have good approaches to prevent protein aggregation, but for peptides not so much. You can make the small step forward that could be the giant leap for mankind. Both biophysical (how do peptides aggregate?) and formulation (how can we stop it?) projects can be designed. Internal project, possibly in collaboration with a company

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No, not a mistake! Much can happen in a year, so who knows what great ideas we get later on. I am also open to suggestions from you, where we can perhaps put a project together that suits your interests and expertise best. Collaborations with other supervisors at the Department can also be designed. Internal project

Master project in industry or abroad
I have many peptide/protein formulation-relevant contacts in the industry (e.g., Zealand Pharma, Novo Nordisk, Ferring), and also abroad (e.g., University of Colorado, Leiden University, University of Groningen, Utrecht University, Ludwig Maximilians University Munich, University of Rome Tor Vergata). So, if this is of interest, come by and have a talk. Note that restrictions will apply, that industrial projects are often defined quite late, and that foreign stays require a lot of work from your side (finding funding being just one of them). But the gain can be substantial!
Drug Delivery of Peptide and Proteins – Design and Biobarriers

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Research Areas
Drug delivery systems design
Oral peptide delivery
Antimicrobials for respiratory tract/skin
Assemblies, nanogels, particles, fibers

Main Experimental Techniques and Methods
Cell culture mucosal models
In vivo models
Bacteria models
Confocal and electron microscopy
Membrane interaction
Mucus and biofilm diffusion
Toxicity assays
Structure/stability assays

Nanogel delivery systems for improved delivery into and across the intestinal mucosa
Aim: Assess how biopharmaceuticals, e.g. insulin, can be encapsulated in polymeric nanogels and co-administered with peptide excipients to improve its delivery across the barriers in the intestine.

Studying the membrane interaction of cell-penetrating peptides with mammalian cells
Aim: Studying the interplay between cell-penetrating peptides and junction-specific peptide excipients with mammalian cell models originating from a variety of human tissue.
Investigating bio-nano interaction for rational design of novel nanomedicines
Aim: Understanding how nanomaterials interact with biological matrices (e.g., mucus and bacterial cell membranes), which is critical for designing effective and safe nanomedicines.

Co-delivery of antimicrobial peptides and conventional antibiotics using biomimetic nanoparticles
Aim: Combating the bacterial biofilm infection by co-delivery of antimicrobial peptides and conventional antibiotics into bacterial biofilm using biomimetic nanoparticle.

Contact: Prof. Hanne Mørck Nielsen (hanne.morck@sund.ku.dk), Assistant Prof: Feng Wan (feng.wan@sund.ku.dk), Post docs: Stine Harloff-Helleberg (stine.harloff@sund.ku.dk), Ditlev Birch (ditlev.birch@sund.ku.dk), Sylvia Kłodzinska (sylwia.klodzinska@sund.ku.dk)
Drug Delivery, Nanomedicines and Radiopharmaceuticals

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For a list of publications and details of our lab, see:
http://www.magneticmicrosphere.com/hafeli_lab/

Research Areas
Targeted Drug Delivery
Nanomedicine Synthesis and Testing
Nanoparticles, Microspheres, Liposomes
Antibodies, Aptamers, Peptides

Main Experimental Techniques
Cell Culture and In Vivo Tests
SPECT/PET/CT Imaging
Scanning Electron Microscopy
Radiolabeling and Quality Control
Toxicity Assays
Protein Assays, Gels
CyTOF, FACS, Confocal
Chemical Conjugations

Please find below examples of typical MSc projects. As all projects require a few months of training in the techniques used, we will only take students for a full 12 months. All these MSc projects will take place in Vancouver, BC, Canada. We will gladly give you the names of previous MSc students, so that you can find out from them what to expect. For more details, please contact us directly!

Biodistribution of a Therapeutic Antibody In Vivo
More and more therapeutic antibodies are being developed for the treatment of many different diseases. Their most common application are in cancer therapy, where they can elicit an effect by themselves, or through a cytotoxic drug or a radioactive isotope, delivered with high efficiency to the target tissue. We typically make the antibody of interest radioactive, measure its stability and cell binding in vitro, and then determine in vivo (mainly by SPECT/PET/CT imaging) its biodistribution and potential toxicity. You as the student will learn all techniques needed.

Development of Less Toxic but More Effective Antibiotics
Many antibiotics are toxic at effective concentrations. We are interested in delivering the antibiotics as nanomedicines that don’t have side effects, for example encapsulated in nanoparticles and microspheres, or bound to polymers. This requires the formulation and optimization of the nanomedicines, their testing in vitro (in minimum inhibitory concentration determinations) as well as in vivo (abscess model). If found effective, biodistribution studies will follow. You as the student will learn all techniques needed.
Testing of Novel Excipients to Allow for Oral Delivery of Biopharmaceuticals

Many biopharmaceuticals, such as insulin and therapeutic peptides, would benefit from being able to be taken orally, as many patients fear injections. We are investigating new polymer- and lipid-based excipients that might allow for the oral delivery of drugs. To determine the fate of the drug and the excipient separately, we make both radioactive and/or fluorescent and then follow them both at the same time in vitro and in vivo. The main technique to be used is diagnostic imaging by SPECT/PET/CT.
Pharmaceutical Physical and Analytical Chemistry Group

The main purpose of the Pharmaceutical Physical and Analytical Chemistry (PPAC) Group is to develop novel approaches for physical chemical characterization of drugs and delivery systems. The work paves the way for design of effective novel medicines and a better understanding of the fate of drug substances and delivery systems through application of quantitative analytical methods.

The PPAC group applies physical chemical approaches in combination with advanced analytical techniques (ICP-MS, SAXS, UV-Imaging and Taylor dispersion analysis) to advance understanding of fundamental processes in drug design, development and characterization. Common to the activities is a focus on the interplay between basic physical chemical properties of drug substances and excipients, kinetics and transport processes in relation to both drug delivery and analytical methods.

**Keywords**: analytical chemistry, cubosomes/hexosomes, drug delivery, excipients, injectables, in vitro release, LC-ICP-MS, metallo-drugs, nanoparticles, preformulation, SAXS, Taylor dispersion analysis

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University of Copenhagen  
Master Thesis, Department of Pharmacy 2018
Jesper Østergaard, Associate Professor
(jesper.ostergaard@sund.ku.dk)

Research interests:
Physical chemical characterization of drug substances and delivery systems.
Development of methods for physical chemical characterization, transport studies, in vitro release and dissolution testing (e.g., UV imaging, capillary electrophoresis (CE), Taylor dispersion analysis (TDA)).
Molecular interactions. Study of non-covalent interactions of drug substances and development of affinity methods based on affinity CE and TDA.
Characterization and development of drug delivery systems for cutaneous administration.
Development of parenteral depot formulation principles (intra-articular and subcutaneous administration), e.g., for treatment of osteoarthritis.
Kinetics. In relation to drug transport processes as well as chemical kinetics and stability testing.

Characterization of nanoparticulate drug delivery systems
Nanotechnology is widely used in the development of new drug therapies. However, significant challenges related to characterization and development remains. This project focuses on the development of new methods to characterize nanoparticulate drug delivery systems, e.g., liposomes, cubosomes, nanocrystals. Traditional analytical chemical procedures are not well-suited for characterization and analysis of these drugs. New micromethods based on capillary electrophoresis, Taylor dispersion analysis and UV imaging will therefore be developed. It is of paramount importance that the methods require limited amount of sample as the new nanoparticulate drug delivery systems are usually only produced in small quantities. The methods will allow us to address critical parameters such as incorporation efficiency, release, non-covalent interactions, aggregation and adsorption.
Supervisors: Jesper Østergaard, Henrik Jensen and Bente Gammelgaard

Novel in vitro drug release models for predicting in vivo performance of injectables
The aim of the project is to develop novel in vitro release testing methods suited for predicting the in vivo fate of injectables. Such methods are of importance in the development of future drugs for subcutaneous, intramuscular or intra-articular injection. We intend to combine a thorough understanding of the transport processes occurring at the injection site with efficient characterizing techniques for unravelling drug release mechanisms and predicting the biological fate. The ultimate validation will be the establishment of an in vitro in vivo correlation (IVIVC).
Supervisors: Jesper Østergaard, Susan Weng Larsen and Henrik Jensen

Compound screening in early drug development using real-time surface dissolution imaging
Surface dissolution imaging provides new opportunities for visualization and study of drug dissolution mechanisms. The aim of the project is to identify and establish best practices in UV imaging-based dissolution testing. The work will involve development of new UV imaging methods for selected test compounds. The project may involve an internship at external partner.
Supervisor: Jesper Østergaard

Development and characterization of NSAID prodrugs for managing joint pain and inflammation
Supervisor: Jesper Østergaard and Susan Weng Larsen
Henrik Jensen, Associate Professor (henrik.jensen@sund.ku.dk)

My research is mainly focused on developing new analytical methodologies for characterization of drugs and drug formulations. The primary focus has been on miniaturization as well as automation of analytical protocols. We often rely on capillary based approaches, but recently we are further developing the methods into wearable sensors for monitoring drugs and their unwanted side effects. Most of my former master thesis students are employed in the pharmaceutical industry, amongst others in the spin-out company FIDA-Tech which is based on research in the group.

FIDA: A new approach for biomarker quantification and immunogenicity assessment

Protein based biomarkers may be used to determine the most optimal drug treatment as well as to monitor treatment status. Therapeutic monitoring of drug compounds is in many cases also beneficial for a successful treatment and recovery. In this project Flow Induced Dispersion Analysis (FIDA) will be investigated for biomarker and immunogenicity assessment. The project may involve internal as well as external collaborations.

Supervisors: Henrik Jensen and Jesper Østergaard

Management of Biopharmaceuticals and Immunogenicity using Wearable Devices

Wearapeutics are wearable devices that can detect biomarkers, monitor and/or deliver a medical treatment and therefore can be the central component of an individualized therapy. At present, the cross disciplinary solutions necessary to realize a widespread application of wearapeutics in individualized therapy are in their infancy and true wearables have only recently been demonstrated for simple sensor systems. In this project, we aim to develop new sensing technologies for biologics (protein based drug compounds) to be used in wearables. The new approaches will be compatible with the small volumes extracted from the subcutaneous environment and will thus rely on capillary electrophoresis, Flow Induced Dispersion Analysis (FIDA) and electrochemistry. The new methodologies will be tested using an in vitro model of wearapeutics for subcutaneous administration. Ultimately, the developed solutions will be adaptable for use with wearable electronics such as smartphones or smart watches.

Supervisors: Henrik Jensen and Jesper Østergaard

Stability assessment of protein based drugs

In this project, the FIDA methodology (see project above) is utilized for stability assessment of protein based drugs. While protein based drugs have proven efficient for the treatment of a range of serious diseases, a number of challenges remains in developing and formulating these drug compounds. Notably, they are known to be structurally labile and efficient methods for assessing stability are currently suboptimal. In this project we take advantage of the fact that structural changes can be monitored as size changes, change in optical properties and altered function (binding ability).

Supervisors: Henrik Jensen
Characterization of exosomes by capillary based approaches
In this project we shall develop a new approach to characterize drug carriers and large MW drugs. In particular we will focus on vesicles and the relatively newly discovered exosomes and their potential as drugs and biomarkers. Exosomes are functional cell derived vesicles with size between 30 and 200 nm making them challenging to study using current methodologies. We will adapt capillary based approaches such as Taylor dispersion analysis, flow induced dispersion analysis and SAXS for studying exosomes.
The project may involve internal as well as external collaborations.
Supervisors: Henrik Jensen, Anan Yaghmur and Jesper Østergaard.
Research Interests: 1) Development of soft nanocarriers based on cubosomes, hexosomes, and other related nanodispersions of inverse non-lamellar liquid crystalline phases. 2) In situ formation of parenteral dosage forms with tunable liquid crystalline nanostructures. 3) Development of cancer nanomedicines based on cubosomes and hexosomes. Of particular interest is the characterization of these nanocarriers by various techniques including SAXS, cryo-TEM, and NTA. In addition, it is also of my interest to design theranostic nanocarriers based on cubosomes and hexosomes.

Current projects:

BRAIN-PENetrating cubosomal and hexosomal NANOcarriers for glioma-targeting delivery. This project focuses on the formulation of immune-safe non-lamellar liquid crystalline nanocarriers for bioimaging of - and concomitant drug delivery to - cancerous brain tumors.

**Supervisor: Anan Yaghmur**

Cubosomes and hexosomes as novel nanocarriers for loading anticancer drugs. The formation/characterization of nanocarriers based on cubosomes and hexosomes for delivery of anticancer drugs.

**Supervisor: Anan Yaghmur**

Microfluidic platforms for the production of monodispersed cubosomes & hexosomes

Combining SAXS to a microfluidic device for the structural characterization of monodispersed drug nanocarriers based on lipid nanoparticles.

**Supervisors: Anan Yaghmur, Aghiad Ghazal**

Cubosomes and hexosomes as antimicrobial peptide nanocarriers

Cubosomes and hexosomes for delivery of anticancer drugs.

**Supervisors: Anan Yaghmur, Stefan Salentini (external Supervisor)**

New stabilizers for cubosomes and hexosomes.

There is great interest to evaluate the use of new biocompatible stabilizing agents.

**Supervisor: Anan Yaghmur**
Self-assembled liquid crystalline nanostructures as sustained release injectable formulations.
The use of non-lamellar liquid crystalline phases as drug delivery systems for intra-articular or subcutaneous administration appear attractive due to the sustained release capability. Combination of biophysical investigations with in vitro release studies.
Supervisors Anan Yaghmur & Susan Weng Larsen

Cubosomes and hexosomes based on omega-3 monoglycerides.
Formation and characterization of lipidic nanoparticles for delivering omega-3 fatty acids (or their combinations with therapeutic agents)
Supervisor: Anan Yaghmur

Selected recent publications:

1. Azmi et al. 2015 (Ther. Deliv.)
2. Azmi et al. 2018 (Langmuir)
3. Yaghmur et al. 2017 (Langmuir)
SUSAN WENG LARSEN

RESEARCH FOCUS: Investigation and application of physicochemical approaches to improve the understanding of processes in drug delivery including parenteral depot design, profiling of drugs and analytical approaches for in vitro release testing. The research is focused on i) development of parenteral sustained release drug delivery systems including in situ suspension-forming injectables, in situ formed liquid crystalline nanostructures and prodrug approaches and ii) development and utility of in vitro release methods to predict in vivo performance of depot formulations for the subcutaneous and intra-articular route of administration.

Local and sustained co-delivery of drugs to improve outcomes following joint replacement
To improve pain management and prevent infections related to joint replacement surgery, there is a need for efficient drug delivery systems (DDSs) that can maintain and sustain an optimal therapeutic level of multiple drugs at the surgical site. The current project focuses on rational design of parenteral in situ forming DDS based on lyotropic non-lamellar liquid crystalline phases for co-delivery of drugs. The drug substances are incorporated in lipids, which self-assemble upon contact with the tissue fluid to form well-ordered inverted type nanostructures (e.g. bi-continuous cubic and hexagonal phases). The aims of the project are to i) study the incorporation of model drugs with different physicochemical properties in these systems and ii) investigate drug release characteristics from in situ formed liquid crystalline phases with various nanostructures. Various in vitro release methods as well as the presence of biologically relevant fluid such as synovial fluid will be used to characterize events influencing the sustained release properties.

Supervisors: Susan Weng Larsen, Anan Yaghmur and Jesper Østergaard

A novel prodrug principle to achieve localized and sustained NSAID action after joint injection
Orally administered NSAIDs are intensively used in the treatment of osteoarthritis for pain management and reduction of inflammation. However, oral NSAIDs may evoke severe side effects even after short term use which constitutes impediments to their use. Following direct injection into the injured joint, a high therapeutic NSAID concentration can be accomplished while minimizing systemic adverse effects. As NSAIDs disappear rapidly from the synovial space (half-lives of 0.5-6 h), depot strategies are needed. In the current project, a prodrug-based in situ suspension-forming drug delivery system intended for localized and sustained NSAID action upon intra-articular injection will be investigated. The project relates to characterization of synthesized NSAID prodrugs and will include: (i) solubility in aqueous buffers and in synovial fluid, (ii) pH dependent stability (pH-rate profile), (iii) sensitivity to enzymatic cleavage (plasma and synovial fluid), (iv) affinity to proteins and (v) rate of NSAID release from prodrugs using an in vitro release model simulating the joint environment.

Supervisors: Susan Weng Larsen and Jesper Østergaard
Design of in vitro drug release models for predicting in vivo performance of depot injectables

Development of in vitro release models for quality control as well as formulation design purposes is a critical activity in the characterization of parenteral depot formulations. Ideally, an in vitro-in vivo correlation should be established, however, it requires that the drug release mechanism is the same in vitro and in vivo. The project focuses on characterizing drug release from sustained release formulations for subcutaneous and/or intra-articular administration. The aim of the project is to develop in vitro release models to achieve in depth understanding of how formulation designs physiological parameters influence the (i) drug release mechanism and rate and (ii) drug transport to the blood capillaries after subcutaneous and/or intra-articular injection. The experimental work will encompass development of release models including column-based continuous-flow systems, dialysis membrane methods and UV-imaging technology.

Supervisors: Susan Weng Larsen, Jesper Østergaard and Henrik Jensen
Huling Mu, Associate Professor

Research focus: Functional excipients and formulation strategies, solid lipid particles and sustained drug delivery, local mucosal drug delivery.

Excipients and drug delivery
Excipients play an essential role in drug delivery. The project aims at exploring the application potentials of functional excipients in regulating drug release and improving drug stability and absorption via a better understanding of interactions between excipients or between drug molecules and excipients. Dosage forms will be selected based on drug molecules (poorly-water soluble drugs or biomacromolecules) and administration route (oral, subcutaneous, or local application). Both lipids and polymeric excipients will be used in formulations, their effects on drug stability and drug release will be investigated systematically using design of experiment.

Supervisor: Huling Mu

Advanced drug delivery by solid lipid particulate systems
Lipid matrix particles can increase the solubility of poorly water-soluble drugs by either increasing the drug solubility or by stabilizing an amorphous or molecular form of drug molecules. The project aims at elucidating the effects of polymorphic forms of lipids on the stability of lipid matrix particles in order to use them as stable carriers in improving the bioavailability of poorly water-soluble drugs for systemic or local therapeutic applications.

Supervisors: Huling Mu and Mingshi Yang

Formulation strategies for sustained drug delivery
The project aims at gaining an insight into the potential of solid lipid microparticles (SLM) as carriers for sustained drug delivery via subcutaneous route. SLM will be prepared using hot melting, emulsification and probe sonication methods, characterized for their size and drug encapsulation efficiency. The impact of formulation and process parameters on drug release will be investigated using design of experiments with selected in vitro models.

Supervisor: Huling Mu and Susan Weng Larsen

Bioadhesive formulations for local mucosal drug delivery
Solid lipid formulations can be used for sustained drug delivery. Incorporation of functional excipients in the formulations may create extra application potentials and be used to improve local therapeutic effect by adhesion to the mucosal surfaces (e.g. mouth, eye). The project aims at investigating the potential of combining bioadhesive excipients and lipid particles and validating the bioadhesive properties of the formulations in vitro. Nanoparticles, prepared by hot melting and probe
sonication method, will be incorporated into buccal films or hydrogels, hence testing drug encapsulation efficiency, drug release, bioadhesion and ex vivo drug permeation.

**Supervisors: Huiling Mu and Jette Jacobsen**

**Synergetic effect of lipids and polymeric excipients on drug delivery**

The project aims at investigating effect of excipients by changing compositions of tablet formulations with different ratio of lipids/polymers. Hot melting emulsification/hot melt extrusion methods will be used and compared to direct compression method. The impact of formulation and process parameters on the characteristics of the tablets will be investigated using design of experiments.

**Supervisors: Huiling Mu and Natalja Genina**

**External projects**

Projects such as oral delivery of proteins can be carried out in collaboration with industry. Please ask for further information.

**Supervisor: Huiling Mu**
Antibacterial Nanoparticles - characterization of nanoparticulate drug delivery systems

Among the most promising novel antibiotic agents are metal nanoparticles, which have shown strong antibacterial activity in numeral studies. Ag is the most commonly applied nanoparticle metal owing to its ancient use as antibacterial agent, but other metals like Au, Cu, Fe, Si, and Ti are frequently used. Owing to their large surface to volume ratio, nano-materials possess distinct physicochemical properties, which influence their effect in the organism. The most important characteristics are composition, concentration, size and size distribution, protein interaction, surface charge and release of ions from the metal.

The focus of these projects is determination of size and concentration by single particle inductively coupled plasma mass spectrometry (SP-ICP-MS). Standards of selected model nanoparticles with different physicochemical properties are used to develop and validate the SP-ICP-MS technique. The methods are applied to authentic nanoparticulate systems received from collaborators. Each project will focus on determination of size and size distribution in standards and quantification in human plasma of a particle system chosen among Ag, Au, FeO, Se, Si, and TiO₂. The projects are the basis for future research on the interaction of nanoparticles in biological systems.

External Projects (Contact me by mail for short descriptions)

Projects at Department of Forensic Medicine:
- CSI Copenhagen - Development of forensic methods for the analysis of drugs of abuse
- Examination of drugs and drugs of abuse in alternative matrix

Projects at National Research Center for the of Working Environment:
- Method development for detection of organophosphate ester metabolites in urine using LC-MS-MS for biomonitoring studies
- Dermal uptake of PCBs using an ex vivo skin model
- Using silicone wristbands for determination of pesticide exposure among agricultural workers in Uganda (The project must start no later than Feb 2019).
Our general research interest is to develop and apply advanced analytical techniques for exploration of novel drug delivery systems and gain knowledge of the uptake, distribution, and metabolism of inorganic and bioinorganic molecules and drugs in biological systems. To pursue this interest we apply LC-ICPMS, CE, and LCMS.

Master thesis projects:

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Determination of trace element impurities in drug products

based analytical methods to comply with Pharmacopeia requirements for elemental impurities. The project is a collaboration with Lundbeck Pharma A/S.
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Master thesis projects:

**Antibacterial Nanoparticles - characterization of nanoparticulate drug delivery systems**

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Physiological Pharmaceutics

In the Physiological Pharmaceutics group (PhysioPharm) we are working within the field of oral and oromucosal drug delivery. We develop novel drug delivery systems (DDS) to improve drug bioavailability and/or to ensure that we get the maximal therapeutic effect.

One example is poorly soluble drugs, where focus is on lipid-based DDS, as well as amorphization, another is peptide and proteins. Further, we also develop formulations for special populations, like children and the elderly.

For the development of delivery systems, we use in vitro models of the mouth and the gastro-intestinal (GI) tract that we are continuously improving in order to make them as physiologically relevant, and predictive, as possible. In addition, we also use and develop in silico model to further understand the absorption and metabolism of the drug after oral administration.

See website for Physiological Pharmaceutics Group

Our work can be divided into the following area:

Mucosal drug delivery
Mucosal drug delivery is a means to improve drug bioavailability and circumvent side-effects. Oromucosal administration is delivery of drugs directly into the oral cavity. It is an attractive route for both local therapy and systemic delivery of drugs, which inherently exhibit variable bioavailability after oral dosing, or low bioavailability due to high first-pass metabolism in the gastro-intestinal tract, liver or skin. Further, this route allows very fast onset of drug action compared to e.g. the oral or the cutaneous route.

Ocular drug delivery is for local treatment of disorders (e.g. dryness due to additional medication) or ocular illness per se.

The research is based on 1) design and characterization of formulations for (new) potential drug substances or 2) gaining mechanistic understanding of clinical observations to develop improved formulations of current drug substances.

In our research, we use validated physiological relevant in vitro and ex vivo methods to evaluate epithelial sensitivity (i.e. safety), drug release, bioadhesion, wettability, and mucosal permeability.

Predictive in vitro models for oral drug delivery
By the use of in vitro models simulating the drug behaviour in the GI tract, from the mouth
to the stomach and further to the lower ileum, we aim at identifying the DDS that will result in the desired therapeutic profile. We aim at understanding the in vivo processes that lead to drug absorption, mainly for small molecules, but also for peptides and proteins. We continuously improve and optimize our in vitro models in order to simulate the relevant GI processes, e.g. release, digestion, partitioning and absorption.

**Lipid-based DDS for oral delivery of poorly water-soluble drugs and peptide/proteins**

Lipid-based DDS (LbDDS) are used in oral delivery of poorly water-soluble drugs by pre-disolving or dispersing drugs in LbDDS. By selecting suitable lipid excipients, certain drugs can be transported via the lymphatic path-way to avoid first pass metabolism. LbDDS is also relevant for oral delivery of proteins and peptides due to their ability to protect proteins and peptides from degradation in the GI tract and the potential incorporation of permeation enhancers. We are especially focusing on self-nano-emulsifying drugs delivery systems (SNEDDS); these are dosed orally as oil in a capsule and will form a drug-containing nano-emulsion in the stomach. SNEDDS can generate a supersaturated drug concentration in the GI tract, which will further promote absorption of poorly soluble drugs. We are also working on incorporating peptides or proteins into SNEDDS, to protect during GI passage and enhance absorption. We are testing the developed LbDDS – and other formulations, in our in vitro models and also in vivo in animal models.

**Drug Delivery Principles**
The above mentioned DDS are often developed as to contain excipients that functionalize their mode-of-action. Bio- or muco-adhesive DDS or mucus penetrating DDS, have gained considerable interest as a means to prolong the residence time of drug to enhance bioavailability. Further, several excipients can be used to enhance permeation of drugs, e.g. medium chain fatty acids, polyarginine and chitosan, or inhibit efflux transporters, e.g. ritonavir or cremophor.

**In silico models for predicting oral drug absorption**

We are working with physiologically-based pharmacokinetic (PBPK) modelling, such as GastroPlus and the SimCyp model. We use the models to understand the relation between release profiles, absorption and the plasma profile and are also optimizing the PBPK models when relevant.

**Note:** As a general rule, a co-supervisor will be allocated to each student, often a PhD student from the PhysioPharm Group, but it can also be a person from the industry or from other institutions.
Supervisors

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Jette Jacobsen, Assoc.Prof., Cand.Pharm., PhD.

**Research focus:** Mucosal drug delivery, oromucosal (buccal, sublingual), bioadhesion, ex vivo permeation ocular, liquid-semi solid- solid formulations, xerostomia

**In vitro in vivo correlation of oromucosal bioadhesiveness -**

of selected polymers with different physico-chemical properties (charge, molecular size, chemical structure) aiming at a platform for screening of oromucosal bioadhesiveness. Today, human in vivo studies of oromucosal bioadhesion are sparse. Examples of qualitative and quantitative in vitro bioadhesion methods to be validated in this project: Zeta potential, tensile strength, rheology, tissue-based retention model, turbidity. Initially, an application to authorities for study of human in vivo oromucosal bioadhesiveness must be written.

**Supervisor:** Jette Jacobsen

**Rule of ## for buccal and sublingual drug absorption to systemic delivery**

This project aims at elucidating the optimal physico-chemical properties of drugs for buccal or sublingual absorption. A series of drugs will be evaluated with regard to solubility in saliva and permeability (cell cultures or ex vivo animal mucosa in side-by-side diffusion cells (e.g. sublingual or buccal mucosa). The project may be planned in collaboration with pharmaceutical industrial partner

**Supervisor:** Jette Jacobsen

**Safety and enhancement of oromucosal or ocular drug permeability**

These projects aim at evaluating excipients (e.g. absorption enhancers, bioadhesive polymer, co-solvents, buffering pH, surface active preservatives, flavours ect.) to gain a mechanistic knowledge of in vitro mucosal permeability of drugs with different physico-chemical properties by employing physiological relevant transport studies with cell cultures or ex vivo animal mucosa in side-by-side diffusion cells (e.g. sublingual or buccal mucosa, and cornea). Potential formulations are liquids (e.g. original product and generics), drugs printed on bioadhesive films (for individualized personal medicine) or solids. The projects may be planned in collaboration with pharmaceutical industrial partner and Chr. Janfelt to perform high-resolution MALDI mass spectrometry imaging of drugs and excipients.

**Supervisors:** Jette Jacobsen, Miriam Kolko (ILF), Susan Weng Larsen, Natalja Genlina, Christian Janfelt

University of Copenhagen

Master Thesis, Department of Pharmacy 2018
Bioadhesive formulations for alleviation of xerostomia

Xerostomia (dry mouth) can be a very disabling state of health. Xerostomia can be due to e.g. use of medicine or illness (Sjögrens syndrome, radiation therapy in cancer). We hypothesize that each, salivary proteins and lipids, contribute to maintain functions of saliva in bulk and at the mucosal surface. The projects will focus on formulation of a physiological relevant composition for daily care and as a drug delivery system. The work may comprise preparation and characterization of semisolid or liquid bioadhesive formulations with and without drugs, development of in vitro cell culture based methods for oromucosal spreadability/wettability (i.e. measurement of single-sided contact angle and surface tension) or friction of formulations, characterization of bioadhesiveness (retention time and force), and mucosal hydration rate. The projects are in collaboration with an external clinical specialist in oromucosal medicine and pathology.

Supervisors: Jette Jacobsen, Huiling Mu and Daniel Bar-Shalom

Bioadhesive lipid particles for local mucosal delivery

Solid lipid formulations can be used for sustained drug delivery. Incorporation of functional excipients in the formulations may create extra application potentials and be used to improve local therapeutic effect by adhesion to the mucosal surfaces (e.g. mouth, eye). The project aims at investigating the potential of combining bioadhesive excipients and lipid particles and validating the bioadhesive properties of the formulations in vitro. Nanoparticles, prepared by hot melting and probe sonication method, will be incorporated into buccal films or hydrogels, hence testing drug encapsulation efficiency, drug release, bioadhesion and ex vivo drug permeation.

Supervisors: Jette Jacobsen and Huiling Mu

External projects

Projects on development and characterisation of formulations can be carried out in collaboration with pharmaceutical industry, Region Hovedstadens Apotek or foreign Universities. Please ask for further information

Supervisor: Jette Jacobsen
Ragna Berthelsen, Assistant Prof., Cand.Pharm., PhD.

Research focus: Design and development of in vitro models to evaluate oral drug performance. In vivo bioavailability studies and predictive in vitro models, simulating the gastro-intestinal tract to understand mechanisms of drug solubilization and absorption. Special focus on drug delivery to the pediatric population.

Project examples:
Development a combined gastrointestinal digestion and permeation in vitro model. Pharmaceutical drug development is a very time consuming, and expensive process. One way to reduce the drug development period and expenses is by generating predictive in vitro models. If well designed, these in vitro model can be used to forecast the relative in vivo performance of various drug delivery systems, thereby ensuring the selection of the best drug delivery system candidates for further studies, minimizing the number of animals studies performed for drug delivery system candidate selection. In order to achieve a systemic response following oral administration, the administrated drug needs to dissolve (or be solubilized) in the gastrointestinal (GI) fluids and permeate the intestinal mucosa to reach the systemic circulation. As many poorly water soluble drugs are affected by the co-administration of food, or formulated in a lipid based drug delivery system to ensure GI drug solubilization, the GI digestion processes will affect the drug absorption. Therefore, two key parameters to evaluate predicting the in vivo performance of orally administrated poorly water soluble drugs are the drug solubilization during GI digestion and the permeation across the intestinal mucosa. As these two processes affect each other, a predictive in vitro model needs to mimic both processes simultaneously. The purpose of the present master's thesis project is to develop and evaluate a combined GI digestion and permeation in vitro model.

Supervisors: Ragna Berthelsen and Anette Müllertz

Lipid based drug delivery systems for the pediatric population
Within the more recent years, there has been an increasing focus on drug delivery to the pediatric population. As the pediatric population differs from the adult population in many aspects, the design of drug delivery systems (DDS) for the pediatric patient requires careful considerations. Oral administration is the preferred route of administration due to the convenience for pediatric patients thus leading to a higher degree of compliance. However, poorly water soluble drugs frequently have a low oral bioavailability. In oral administration, the physiological conditions of the gastro-intestinal tract (GIT) is so complex that the rate and extent of drug absorption are influenced by many factors, especially when the drug solubility is low, it often causes
incomplete absorption and low bioavailability, seriously affecting the drugs clinical efficacy and the treatment of the disease.

The project may involve: 1) construction of a pediatric lipid based DDS, and 2) in vitro evaluation of the designed DDS, studying drug solubilization capacity and mechanism, stability and drug release profiles using in vitro pediatric digestion model, and 3) studies of the permeation of poorly soluble drugs following administration of the designed DDS.

Supervisors: Ragna Berthelsen, Anette Müllertz, and Xioana Lui (phd student)

Is intestinal mucus an important factor in solubilization and absorption of poorly soluble drugs?

Efficient oral therapy is dependent on a good and reproducible drug absorption. However, many active drug candidate suffer from poor water solubility, which makes it difficult to formulate them into delivery systems with high and reproducible bioavailability. One important factor influencing this, is the poor understanding of the actual conditions in the gastrointestinal tract, and the factors that are limiting for the drug absorption. Therefore, the tools that are used in the development process are in many cases not predictive for the in vivo situation. One example is the media that are used for predicting the drug dissolution in the intestine, which often underestimates the in vivo dissolution. We hypothesize that the intestinal mucus layer, has a larger solubilization capacity than the intestinal fluids and is therefore a major player in the absorption mechanism for poorly water soluble drugs. Thus, the mucus layer need to be included in in vitro models simulating drug dissolution and absorption.

This project aims to elucidate the impact of the intestinal mucus layer in dissolution, solubilization, diffusion and absorption of poorly water soluble drugs.

Supervisors: Ragna Berthelsen, Anette Müllertz, and Mette Kiltgaard (Phd student).
Daniel Bar-Shalom, Assoc.Prof., Cand.Pharm.

Research focus: Pediatric and geriatric formulations. Functional excipients. Implementing learnings from food science and material science into pharmaceutical science.

Development of “instant pudding” vehicles
There are different scenarios requiring different solutions: (a) Individual, single dose commercial products to be produced by the industry, (b) Vehicles to be added to magisterial compounded products at the hospital or community pharmacy and (c) Vehicles for mass treatment in the developing world where a whole village is treated at once (for example, with anti-parasitic combinations) etc. The projects are usually carried out in collaboration with interested parties (pharmaceutical and food industry, raw materials suppliers, analytical instrumentation producers and academic partners. Many of those abroad)
Supervisor: Daniel Bar-Shalom

Paediatric/Geriatric drug delivery.
Children and the elderly are special population groups from the oral drug delivery perspective. Their swallowing (in-) abilities are different from those of the “average population” (those able to swallow tablets and capsules). The elderly and chronically sick children tend to take multiple drugs, thereby complicating the treatment. Our approach is to individually microencapsulate the drugs to eliminate the (bad) taste problem, to prevent unfortunate interactions between the drugs when in combination and to provide controlled release, if possible. The microencapsulates are mixed with dry, “instant pudding” formulations, and just before administration water is added, resulting in a pleasant, easy to swallow pudding/applesauce mass.
Supervisor: Daniel Bar-Shalom

Development of microencapsulated particles for children and elderly
Different applications require different solutions. The drugs used at the hospital are a very heterogeneous group, those given in anti-parasitic treatments are notoriously water insoluble, in single doses and long-term stability is expected.
Supervisor: Daniel Bar-Shalom

Development of the dispensing systems for Microencapsulate/Pudding products
It is not sufficient to have the components of the product, a dispensing strategy and device are needed too. Project in this part involve collaboration with design experts.
Supervisor: Daniel Bar-Shalom

Oral controlled release of high dose, highly soluble drugs
The problem of insoluble drugs is much researched all over the world, including people in
our own department. The opposite problem has been largely ignored, but it is there and represents an interesting "niche".

Supervisor: Daniel Bar-Shalom

Reduction of water solubility by combination with water soluble polymer

Unexpectedly, it has been found that some water soluble drugs, when melted together with water soluble polymers (normally used to enhance the solubilization of water insoluble drugs) their rate of dissolution is reduced. The projects explore the mechanisms involved and the (potential) practical applications.

Supervisor: Daniel Bar-Shalom
Anette Møllertz, professor, group leader

Research focus: Development of predictive in vitro models, simulating the GI tract. Lipid-based drug delivery systems for oral delivery of both poorly soluble drugs and peptide/proteins. Small-scale methods to evaluate solubility, dissolution and supersaturation propensity of drugs. Application and development of in silico PBPK models to evaluate release effect on pharmacokinetics.

Development of a targeted drug delivery system by use of micro-containers

Inflammatory bowel disease (IBD) is an autoimmune disease that affects more than 50,000 people in Denmark. Current treatment options are not effective and are often associated with severe side effects due to systemic uptake of the drug. A potential oral drug delivery system (DDS) for treatment of IBD is micro meter sized containers (micro-containers). They are small polymeric cylinders that are open in one end, thus allowing filling of API. The open ends are sealed with a suitable polymer depending on the purpose of the DDS. In this way, micro-containers provide protection of the loaded API from both low pH and enzymatic degradation while ensuring unidirectional release of the API in the intestine. The aim is to develop a DDS that targets only the inflamed areas of the gastrointestinal tract in patients suffering from IBD. This can be achieved e.g. by functionalization of the polymer lid and/or targeting ligands. Newly developed DDS will be tested in vitro, and the promising DDS will be investigated in vivo in a preclinical IBD rodent model.

Supervisors: Anette Møllertz, Thomas Rades & Maja Nørgaard Kristensen (PhD student)

The effect of gastric transfer rate on supersaturation and precipitation of poorly water soluble weak bases (60 ects)

A large solubility difference between the low pH stomach and neutral pH small intestine is a challenge for many novel drug compounds. The large difference in solubility can lead to supersaturation and subsequent precipitation in the small intestine, which may induce variability in fraction absorbed. This can lead to a large intersubject variability in pharmacokinetic studies. Supersaturation is an unstable system, where the concentration of a compound exceeds the (thermodynamic) solubility and over time will precipitate to give a concentration corresponding to the solubility.

The aim is to establish a novel two-step dissolution model “the BioGIT”, with a gastric and an intestinal compartment to investigate the effect of gastric transfer rate on small intestinal supersaturation and precipitation of different formulations of poorly water soluble weak bases. The project will include a 3-6 month stay in Professor Christos Reppas lab at National and Kapodistrian University of Athens, Greece, to learn the method and then set it up in the PhysioPharm group. Different formulations will be tested, HPLC will be used to quantify the drug, while XRPD, Raman or DSC will be used to determine the solid form of the drug.

Supervisors: Anette Møllertz & Jakob Plum
Simulating food effect on poorly soluble drugs

Many poorly soluble drugs have positive food effect, meaning that they have a better bioavailability in the fed state. This can be a problem for the reproducibility of the therapeutic effect of the drug and it is therefore desired to develop formulations that abolish the food effect. The aim of this project is to develop in vitro models, simulating the fed state GI tract, and by use of this to develop formulations without food effect.

Supervisor: Anette Müllertz

Functionalized self-emulsifying DDS (f-SEDDS) for oral delivery of peptides or proteins

Self-emulsifying drug delivery systems (SEDDS) are isotropic mixtures of oil, surfactant and co-surfactant spontaneously forming an oil/water emulsion upon mixing with an aqueous medium. SEDDS have shown great potential to improve the bioavailability of peptide/protein (P/P) drugs by having advantages as mild preparation conditions, protection against enzymatic hydrolysis and build-in permeation enhancement. Pickering emulsions, wherein solid colloidal particles stabilize the emulsion system by adsorbing at the oil/water interface, present advantages in terms of stabilizing the peptide, while incorporation of muco-adhesive polymers, like chitosan, to the SEDDS, could be an attractive alternative to increase residence time in the intestine. The project will include the development of solid colloidal particle (i.e. silica or graphene oxide) functionalized SEDDS (f-SEDDS) and/or mucoadhesive f-SEDDS. F-SEDDS will be characterized by particle size, zeta-potential and in vitro digestion. The performance of the developed f-SEDDS will be evaluated by in vitro cell culture study and in vivo animal models.

Supervisor: Anette Müllertz

Supersaturated self-nano-emulsifying DDS (super-SNEDDS) for oral delivery of poorly soluble drugs

Previously we have shown that some drugs can form a stable supersaturated state in a SNEDDS and that this can result in improved bioavailability. In this study we want to find the maximum degree of supersaturation and also if polymers and amorphous drugs can further increase this. Further we want to understand the mechanisms behind, especially with regard to the mechanism of absorption.

Supervisors: Anette Müllertz and Thomas Rades

Assessing the relation between release profile and pharmacokinetics using PBPK models

Physiologically based pharmacokinetic (PBPK) modelling is important in the drug development process to understand the relation between drug release, absorption and plasma profile and thereby to select the drug candidate and formulation approaches. However, based on our experience, there is still shortcomings in the input (absorption) profiles in the PBPK models, e.g. the entire drug solubility profile in the GI tract is not taken into account. In this project we want to apply PBPK models to different solubility and dissolution profiles, from selected formulations. We will use the softwares GastroPlus and SimCyp.

Supervisors: Anette Müllertz, Ragna Berthelsen, Jakob Plum
Solid State Pharmaceutics Group

We develop drug delivery systems based on high-energy solids. The aims are improved stability of the formulations, and solubility enhancement of poorly water-soluble drugs. The scientific staff consists currently of the three scientists Korbinian Löbmann, Thomas Rades and Holger Grohganz, and a varying number of PhD students. The research in the group is currently covering two main areas:

Amorphous drug formulations

By stabilizing a poorly soluble drug in its amorphous state, its bioavailability can be increased, due to a higher dissolution rate and solubility. The amorphous drug can be stabilized with both polymers and small molecules. The amorphous drug and its stability is characterized by a state-of-the-art methods. Typically amorphous drugs are dosed in tablet, and we work on functionalizing the tablets as to increase permeation or inhibit efflux transporters. Low solubility is the major challenge for future small molecule drugs. Overcoming the inherent instability of amorphous formulations is one solution to the solubility problem.

Freeze-dried biopharmaceutical formulations

Process and formulation parameters influence the solid state of both excipient and macromolecule, and thus quality of freeze-dried protein formulations. Understanding the interactions and solid state properties of a macromolecular formulation is important for the design of a stable formulation.
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Projects

Quality by design (QbD) in the processing of biomacromolecules

The quality by design (QbD) principle can be expected to influence the way of pharmaceutical processing in the years to come towards the development of more rational processes. Although, freeze-drying and spray-drying are widely used in peptide and protein formulation, the interaction between various excipients and proteins is not fully understood. It is the aim of the projects to obtain a deeper understanding of the influence of various composition and process parameters on the solid state form of both the excipient and the macromolecule. Analytical techniques may include X-Ray powder diffraction, dynamic mechanical analysis, NIR and Raman spectroscopy as well as the application of multivariate data analysis.

Evaluation of preparation methods for co-amorphous formulations

Low drug solubility is the major challenge for future small molecule drugs. In order to overcome the problematic solubility of BC class 2 drugs, small excipients are investigated to form co-amorphous formulations. Due to the low solubility, ball milling is used as preferred production process. Due to the low capacity of ball mills, other production procedures, such as freeze-drying, spray-drying and hot-melt processing should be evaluated. Project in co-operation with Thomas Rades.
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Projects
On the role of water in amorphous systems.
Water is generally seen as detrimental for the stability of amorphous drugs and solid dispersions. However, initial in-house work has shown that this may not necessarily be true for all systems especially at low water concentrations. The aim of this Masters project is to determine the stability of a range of amorphous systems (pure drugs, amorphous solid dispersions with polymers and co-amorphous systems with amino acids) in the presence of different amounts of water. The student will learn preparative techniques, including ball milling and quench cooling as well as using a wide range of analytical techniques, including X-ray powder diffraction, FTIR spectroscopy, thermal analytical techniques, water determination and dissolution techniques. Project in co-operation with Matthias Manne Knopp (suitable for 2 MPharm Students)

On the role of molecular weight and viscosity in precipitation inhibition.
The project involves the characterization of amorphous drug polymer mixtures. In this study we want to investigate the role of polymer molecular weight and viscosity on the dissolution and precipitation inhibiting properties of polymers on a range of model drugs. The hypothesis of this work is that both the viscosity enhancing properties of polymers as well as specific drug polymer interactions play a role in inhibiting the drug precipitation form the supersaturated state. To delineate these two effects, polymers of the same type but different molecular weight will be used at similar molar ratios to the drug, calculated as ratio of drug to repeat unit. If successful, this will be the first study of its kind, and will help elucidating the mechanism of drug precipitation inhibition. We expect a publication in an international peer reviewed journal to result from this work. Project in co-operation with Matthias Manne Knopp (suitable for 2 MPharm Students)

Electrospun amorphous solid dispersions of poorly water-soluble drugs. The development of oral dosage forms from poorly water-soluble active pharmaceutical ingredients (APIs) remains a major challenge for the pharmaceutical industry. Preparing amorphous solid dispersions (ASDs) allows increasing the solubility and dissolution rate of an API, hence, increasing its bioavailability. The application of electrical energy during electrospinning can generate ASD nanofibers from drug-loaded solutions and melts. This project focuses on the development of an electrospinning method to produce ASDs from the corn protein zein and several poorly-water soluble APIs. The APIs will either be electrospun co-axially or directly from a mixture with zein. The electrospun fibers will be comprehensively
characterized by a range of analytical techniques, including scanning electron microscopy, X-ray powder diffraction, differential scanning calorimetry and dynamic mechanical analysis. The dissolution rate advantage will also be determined. Project in co-operation with Andrea Heinz (suitable for 2 MPharm Students)
Projects

**In situ amorphization using microwave irradiation.**

A major concern of amorphous formulations is their physical instability, which puts the gain in solubility at risk upon long time storage. Using the concept of in situ amorphization, microwave activation will transform a poorly soluble but thermodynamically stable crystalline drug into its highly soluble but thermodynamically unstable amorphous counterpart within the final dosage form (tablet) prior to administration. The aim of the project is to identify promising drug-polymer combinations that are suitable for microwave activation and hence develop a new drug delivery system. *Project in co-operation with Nele Hempel (suitable for 2 MPharm Students).*

**Development of novel DES based supersaturating drug delivery system for poorly soluble drugs**

Deep eutectic solvents (DESs) have been used as an environmental-friendly alternative to organic solvents in a wide variety of applications in the chemical industry. DESs are mixtures of two or more non-toxic organic compounds such as amino acids, sugars or carboxylic acids. The solubility of poorly soluble drugs in DES has been reported to be up to 55,000 fold higher than in water. Hence, DESs have been suggested as a potential drug delivery system for poorly soluble drugs. The aim of this project is to identify feasible drug/DES combinations and develop a supersaturating drug delivery system capable of maintaining the drug in the supersaturated state. *Project in co-operation with Henrik Palmelund (suitable for 2 MPharm Students).*
Tiny medicine sponges – Understanding drug adsorption and molecular mobility in mesoporous silica.

Here we want to increase the mechanistic understanding of molecular interactions and mobility of adsorbed drug monolayers on mesoporous silica (MS) surfaces. These include the impact of physico-chemical properties of the adsorbed drugs and MS-drug molecular interactions on the monolayer loading capacity. Furthermore, we want to increase the understanding of the monolayer mobility and phase transition, which we recently were able to experimentally identify.

Lastly, to date there is still no feasible solvent free loading procedure available, which we aim to develop by facilitating the drug adsorption to the MS surface. **Project in co-operation with Matthias Manne Knopp (suitable for 3 MPharm Students).**
Surface and Colloid Chemistry Group

The goals of the Surface and Colloid Chemistry Group is to advance the knowledge of interactions of host defense peptides and nanomaterials as carriers for such systems, with membranes and membrane components of cells and bacteria. Through this, as well as innovative approaches for performing such studies, the goal is also to bring the research to a stage where it can be translated to further therapeutic development.

See website for Surface and Colloid Chemistry Group

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Project examples

Membrane Interactions of structured silica nanoparticles as antimicrobial agents
In the wake of increasing bacterial resistance against conventional antibiotics, there is a growing interest in alternative approaches for reaching antimicrobial effects. Among those, nanoparticles are attracting considerable current interest due to the comparatively low cost, good scalability, and broad versatility of such materials, but also due to presently undeveloped bacterial resistance. Here, various nanomaterials offer opportunities for triggered functionalities to combat challenging infections. Although the performance in these diverse applications is governed by a complex interplay between the nanomaterial, the properties of included drugs (if any), and the biological system, nanoparticle-membrane interactions constitute a key initial step and play a key role for the subsequent biological response. Clarifying key factors controlling membrane binding and destabilization of nanoparticles is therefore key for the successful development of the latter towards therapeutics.

In the present project, membrane interactions will be investigated for mesoporous silica nanoparticles, which offer opportunities in combating challenging infections. For example, both low molecular weight and biomacromolecular drugs can be readily incorporated into such nanoparticles, allowing large drug loads due to their large specific surface area. In
addition, silica nanoparticles can be designed to display needle-like surfaces, which may be used to effectively "puncture" bacterial membranes by "needle-like" actions. Within the project, factors determining membrane interactions of "spiky" mesoporous silica nanoparticles will therefore be investigated by previously developed model lipid membranes, in combination with various biophysical techniques, such as QCM-D, ATR-IR, and light scattering. Results from such biophysical studies, e.g., on effects of membrane composition, structure and charge of the silica nanoparticles, and effects of co-administration of such particles with other potent antimicrobial agents, notably antimicrobial peptides, will be correlated to biological results on antimicrobial effects and cell toxicity for selected systems.

**Supervisor: Martin Malmsten**

**Number of students:** 1.

**Membrane Interactions of gold nanoparticles as antimicrobial agents**

In the present project, membrane interactions will be investigated for gold nanoparticles, which offer opportunities in combating challenging infections. For example, both low molecular weight and biomacromolecular drugs can be readily adsorbed at the surface of these nanoparticles, allowing large drug loads due to their large specific surface area. In addition, metal nanoparticles (including gold) display potent antimicrobial effects by themselves. The latter originate from several mechanisms, including direct membrane rupture, binding to sulfhydryl groups of metabolic enzymes, binding to microbial DNA, and generation of reactive oxygen species, in turn causing bacterial enzyme and lipid oxidation. Furthermore, gold nanoparticles may be locally heated by light, which can be used for killing even multi-resistant and biofilm-forming pathogens. Within the project, factors determining membrane interactions of gold nanoparticles will therefore be investigated by previously developed model lipid membranes, in combination with various biophysical techniques, such as QCM-D, ATR-IR, and light scattering. Results from such biophysical studies, e.g., on effects of membrane composition, size and surface properties of the gold nanoparticles, and effects of co-administration of such particles with other potent antimicrobial agents, notably antimicrobial peptides (3), will be correlated to biological results on antimicrobial effects and cell toxicity for selected systems.

**Supervisor: Martin Malmsten**

**Number of students:** 2.
Application of oxidative stress on lipid membranes as a new tool for developing new antimicrobial agents

Aiming to improve the efficacy and safety of novel therapeutics and based on important advances in material science, drug delivery research is currently undergoing considerable growth to include a range of interesting nanomaterials. Particularly, inorganic nanoparticles are very attractive as antimicrobial agents ('nanobiotics'), notably due to the increasing resistance development against conventional antibiotics. Apart from scalability and versatility, such materials offer advantages related to responsiveness of antimicrobial and anti-inflammatory effects and the possibility of controlling them by a range of triggering factors. Inorganic nanomaterials may provide antimicrobial effects through a number of mechanisms, including direct membrane binding destabilization, release of antimicrobial metal ions, generation of reactive oxygen species, or localized heating induced by either light or oscillating magnetic fields. Generally, oxidative stress is known to be a key antimicrobial mechanism for a range of nanomaterials such as TiO₂ nanoparticles, which can be activated by UV light. However, studies on the mechanisms underlying this remain scarce. Based on previous work done by our group on the oxidative destabilization of lipid membranes, and using different biophysical and surface-chemical techniques, the present project will focus on the effects of nanoparticle-induced oxidative stress on lipid degradation, its consequences for membrane structure and stability, and any potential membrane selectivity that could allow more efficient antimicrobial activities along with reduced side effects. In a wider perspective, this project will contribute to the mechanistic foundation for the use of photocatalytic nanomaterials as antimicrobial agents.

Supervisor: Elisa Parra & Martin Malmsten
Number of students: 1.

The effect of electrostatics on the formation and functionality of AMP-loaded microgels

Microgels have been found to be advantageous systems for the delivery of biopharmaceuticals, such as antimicrobial peptides (AMP), due to their triggerable swelling properties and protectiveness against enzymatic degradation. However, studies for the use of microgels as carriers of AMP are still relatively scarce, where the lack of systematic studies considering peptide loading, particle formation and membrane interactions, makes the underlying mechanisms behind the effectiveness of the mentioned systems difficult to understand. In this work, the interactions of peptide-loaded microgels with bacteria like lipid vesicles will be investigated with a combination of techniques, such as fluorescence spectroscopy and QCM-D. For this aim, microgels will be prepared with a microfluidic approach, varying parameters such as pH, ionic strength, and cross-linking degree.

Supervisors: Martin Malmsten & Bruno C. Borro
Number of students: 1
Vaccine Design and Delivery Group

One of the primary goals of the Vaccine Design and Delivery Group is to gain new fundamental knowledge that can facilitate the design, optimization and development of novel delivery systems capable of delivering loaded biopharmaceuticals to the intended target site(s). The group addresses the complex challenges associated with the formulation and targeted delivery of vaccines and nucleic acid-based therapeutics.

The research focus of the Vaccine Design and Delivery Group is advanced drug delivery. The group uses in vivo imaging (e.g., NIR, MRI and SPECT/CT) to guide and optimize the design of new nanoparticle-based delivery systems for vaccines and nucleic acid-based therapeutics to improve therapy. The research in the group is highly interdisciplinary. The team comprises 12 international researchers of various backgrounds, including pharmacists, biologists, biochemists, physicists and biotechnologists.

Biopharmaceuticals like vaccines and nucleic acid-based therapeutics are challenging from a pharmaceutical perspective, because they are highly complex products, which can only be understood via solid fundamental science. Using systematic knowledge-based quality-by-design approaches, combined with advanced physicochemical and biopharmaceutical assessment, and molecular imaging, we design new delivery systems for biopharmaceuticals to optimize their stability, efficacy and safety.

The overall research goal is to improve disease prevention and treatment in the fields of infectious and inflammatory diseases, and cancer. The group is addressing drug delivery challenges from bench-to-bedside, via international collaborations, using state-of-the-art technologies. This has fostered innovative solutions and high-impact publications in drug delivery.

See website for Vaccine Design and Delivery Group

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Nanoparticles for nucleic acid and vaccine delivery
Nanoparticles are highly attractive as delivery systems for nucleic acids and vaccine antigens, because they mimic the structure and/or composition of microbes that are naturally adapted to infect target cells of the host. However, the specific design of nanoparticles is crucial for ensuring optimal delivery of the active pharmaceutical ingredient (API) to the target cells, as well as an acceptable safety profile. A central question for our research is to
mechanistically understand how the design of nanomedicines affects the interaction with the environment, e.g., in formulation, in vitro and in vivo. This basic knowledge is applied to optimize the efficacy and safety of the formulation. We investigate lipid- as well as polymer-based nanoparticulate drug delivery systems for delivery of nucleic acids and vaccines using various approaches for targeting and membrane destabilization. Nanoparticulate delivery systems are highly complex molecular assemblies, hence exhaustive physicochemical characterization is of major importance for gaining an increased understanding of nanoparticle structure-activity relationships. Advanced characterization techniques are used for this purpose. We perform our research in interdisciplinary, collaborative and international projects.

Please find examples of projects in Vaccine Design and Delivery Group on the following pages.

Project examples
Magnetic resonance imaging-assisted design of a thermostable and self-administrable tuberculosis vaccine for inhalation
Tuberculosis (TB) is caused by Mycobacterium tuberculosis, which enters the lungs and other organs through the respiratory mucosa. The objectives of this project are to perform real time quantitative tracking of a pulmonary candidate vaccine for TB, to achieve optimal in vivo dendritic cell targeting and T cell activation for sustained memory T cells, and translate these findings into a safe and improved prophylactic vaccine formulation for TB. Using in vivo molecular imaging system that combines PET with CT or MRI and fluorescence imaging, we will generate ADME profiling of the pulmonary candidate TB vaccine and in vivo track the vaccine components separately. The results will improve our understanding of the safety and efficacy requirements of a pulmonary vaccine candidate for TB in pre-clinical models before clinical trials in humans. A pulmonary delivered TB vaccine can significantly accelerate protective mucosal Th1 immune responses and improve the central memory T cell pool, which is usually exhausted in BCG-vaccinated adults.

Supervisors: Camilla Foged and Aneesh Thakur
Duration: 12 months
Working place: UCPH
Microfluidics-assisted design of next-generation mRNA vaccines – a novel tool for fighting cancers and challenging infectious diseases

Although vaccines have had a tremendous positive impact on global health, they are expected to be even more important in the future due to the increased frequency of drug-resistant microorganisms and diseases representing so-called "difficult targets" against which we do not have effective vaccines yet, e.g., tuberculosis, AIDS and cancer. One promising strategy is the use of vaccines based on mRNA that encode antigenic proteins from pathogens or tumour cells. In fact, mRNA vaccines represent a new era in vaccinology. However, challenges include the effective delivery of mRNA across cell membranes to the immune cell cytosol, and proper induction of cytotoxic T lymphocytes (CTLs) that then can kill infected or transformed cells. Recently, we have made major breakthroughs addressing both of these challenges by identifying important constituents of such vaccines: an innovative delivery system based on nanoparticles that delivers nucleic acids to the cytosol. In addition, we have identified molecular components needed to induce strong CTL responses. In this project, these two principles will be combined into one multifunctional nanoparticle-based vaccine that both delivers mRNA encoding antigen and induces CTL responses. Microfluidics will be used for manufacturing to precisely control the particle properties and to ensure the scalability of the production process, which is essential for translating the concept to the clinic. The efficacy and safety of the mRNA vaccines will be tested in animal models of infection and cancer.

Supervisors: Camilla Foged and Dennis Christensen, Dep. of Infectious Disease Immunology, SSI

Duration: 12 months

Working place: UCPH and SSI

Design and Investigation of nanoparticles for targeted delivery of small interfering RNA (siRNA)

Recently, the use of small interfering RNA (siRNA) has received an overwhelming amount of attention for the treatment of serious diseases such as cancer, infectious and inflammatory diseases. A key hurdle for the further development of RNA interference (RNAi) therapeutics like siRNA is their safe and effective delivery because it has been realized that protective delivery systems are required that can facilitate their intracellular delivery. In this project, we focus on the rational design of nanoparticle-based siRNA carriers and the detailed molecular understanding of the systems in formulation and in biological environments. Master projects can embrace parts of the following ongoing research projects in the group, depending on your specific interests: i) Polymeric nanoparticle and liposome engineering and optimization through detailed physicochemical characterization, ii) processing of nanoparticles
(spray drying and freeze drying), iii) mechanistic aspect of the interaction between nanoparticles and the target cells, including uptake mechanisms, intracellular trafficking and intracellular drug release, and iv) in vivo delivery of siRNA.

**Supervisors:** Camilla Foged  
**Duration:** 6-12 months  
**Working place:** UCPH

**Examples of external master thesis projects**

**Formulation development of vaccines**
The project will be conducted at Janssen Vaccines, Leiden, The Netherlands. The student will be involved in the daily activities of a multidisciplinary team with people of different nationalities and will experience at close-hand the (bio-) pharmaceutical industry. The student will work on the formulation development of a vaccine. This “vaccine X” will need to be stabilized, which might require the addition of an additional immune stimulating component, an adjuvant. Interaction(s) of the antigen(s) with the adjuvant will be investigated. The student will use and develop high throughput technologies (robotic liquid handling in combination with high-throughput analysis) in order to characterize the physical and chemical stability of the vaccine formulations. Typical activities will likely include the development of procedures and assays for formulation development, design-of-experiment (DoE), preparation of formulations, stability testing, sample and data analysis, presentation of results.

**Supervisors:** Camilla Foged and Martinus Capelle, Janssen Vaccines, Leiden  
**Duration:** 12 months  
**Working place:** Janssen Vaccines, Leiden, The Netherlands

**Development and evaluation of micro-environmentally responsive drug and vaccine therapies to treat cancer**
This project will be conducted in collaboration with Professor Sarah Hook at the University of Otago in Dunedin New Zealand. The student will work on a project developing responsive sustained release systems for the targeted delivery of oncology drugs and vaccines. The project will involve developing and characterizing various formulations at the chemical, physical and biological level.

**Supervisors:** Camilla Foged and Sarah Hook  
**Duration:** 12 months  
**Working place:** University of Otago, Dunedin, New Zealand
Master project in The Netherlands (Utrecht)
Did you ever think of doing your master project in the Netherlands? We have close collaborations with excellent groups at Utrecht University that offer challenging research environments with interesting research projects in the field of drug delivery.
For a more general description of the research activities, check their website. Contact me for more information and possible projects.

Supervisors: Camilla Foged and local supervisor
Duration: 6-12 months
Working place: Utrecht, the Netherlands

Photoporation as a novel drug delivery technology
There is great interest in delivering macromolecular agents like siRNA and contrast agents such as quantum dots into living cells for therapeutic and diagnostic purposes. Although substantial effort has gone into designing non-viral nanocarriers for intracellular delivery of these materials, entrapment in endosomes after endocytotic uptake remains a major bottleneck. Laser-induced photoporation in combination with plasmonic (e.g. gold) nanoparticles is an alternative physical method that is receiving increasing attention for delivering macromolecules in cells. By using appropriate pulsed laser light, gold nanoparticles can be quickly heated so that water vapour nanobubbles can emerge in hydrated tissue. When the gold nanoparticles are adsorbed to the cell membrane, these laser-induced vapour nanobubbles can create pores in the membrane. Macromolecules or nanoparticles in the surrounding cell medium can then diffuse through the pores directly into the cell’s cytoplasm. Using this photoporation methodology, we have demonstrated that cells can be efficiently transfected with siRNA, including hard to transfect cytotoxic T-cells. In addition, we have shown that vapour nanobubble photoporation is ideally suited for efficient delivery of contrast agents like fluorescent dextran and quantum dots into cells. Applications range from long-term cell tracking for in vivo cell therapies, to labelling of selected cells (e.g., primary neuronal cells) in culture for high-content screening. Combined with the fact that little or no cytotoxicity is induced, it is clear that laser-induced membrane poration by vapour nanobubbles is highly promising delivery method whose applications have only just begun to be explored. Current research in the group explores various applications of this exciting new technology in the field of drug and nanomaterial delivery.

See website for Gent University.
Supervisors: Camilla Foged and local supervisor
Duration: 6-12 months
Working place: Gent, Belgium

Studying and overcoming bio-barriers in drug delivery
In drug delivery, intensive research is being carried out to develop ‘intelligent’ nanocarriers that are capable of efficiently delivering biopharmaceuticals to target cells. These nanocarriers should fulfil several requirements in order to deliver their therapeutic cargo to
the intended target cells. During the entire delivery process in the body they should be colloidally stable, provide protection against degradation of the cargo, have excellent mobility through extracellular tissues and deliver their cargo in the cytosol or nucleus of the target cells. Obtaining a better insight into the physicochemical and biophysical properties of nanomedicines during the various phases of the delivery process is required to achieve efficient optimization of their structure and composition. In the past decade we have developed several advanced fluorescence microscopy methods that enable detailed investigation of the dynamic interaction of nanomedicines with various biological tissues. Methods have been developed to study colloidal stability of nanomedicine formulations in undiluted biological fluids like blood. Other studies have focused on characterizing nanomedicine mobility in extracellular tissues like cystic fibrosis mucus or vitreous humour. Also, cellular uptake and intracellular processing of nanomedicines has been investigated in great detail.

By providing a better insight into the stability and transport of nanoparticles during the various phases of the delivery process through the use of advanced microscopy techniques, it is our aim to enable a more efficient and rational development of improved carrier materials for the delivery of nucleic acids. See website for Gent University.

Supervisors: Camilla Foged and local supervisor
Duration: 6-12 months
Working place: Gent, Belgium
Manufacturing and Materials

The Manufacturing and Materials group is focusing on processing and material sciences around solid state and semisolid pharmaceuticals. This international group has over 20 PhD students, postdocs and assistant/associate professors. The section is aiming for understanding the chemical and physical properties of the active pharmaceutical ingredients, both small molecules and bio-macromolecules, and excipients in relation to their processing behaviour. Further, the group aims to optimize stability and bioavailability of the final dosage form. The research in group is focusing on facilitating the future implementation of Quality by Design (QbD) principles for drug development processes, as well as implementation of process analytical technologies (PAT) for industrially relevant applications.

We focus on processing of small-molecule and protein/peptide-based medicinal products. Our goal is to understand the properties of active pharmaceutical ingredients and excipients in relation to their processing behaviour, and further, in relation to the optimal bioavailability of the final dosage form.

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Please find examples of project examples in Pharmaceutical Technology and Engineering on the following pages ...
Lene Jørgensen
Associate Professor and Head of Studies Master of Medicines Regulatory Affairs (MRA)

Research focus: protein stability, protein formulation, adsorption to surfaces, manufacturing and processing of protein pharmaceuticals, excipients design and requirements, quality of pharmaceutical products, regulatory aspects of manufacturing pharmaceuticals.

Examples of projects:

- **Hot melt processing of biopharmaceuticals**
  
  In this project, the amorphous environment created by various polymers in a hot melt process will be studied to see whether it is sufficient to prevent the degradation caused by heat, and therefore would enable protein formulations to be produced by hot melt processing. This will be investigated by monitoring structural changes in both the solid and the liquid state. Several process and formulation variables form a good basis for the application of Design of experiments (DoE).

  **Supervisors: Lene Jørgensen and Johan Peter Bøtker**

- **Physical stability of proteins**
  
  Maintaining the physical stability of proteins is crucial in both formulation and manufacturing, since the biological activity may otherwise be reduced. Most formulations containing proteins are simple solutions or freeze-dried products. We will expose a protein to various types of stresses, formulation components, processing and use advanced techniques to characterize the changes that we observed. As a part of the formulation study, we will add different types of excipients and determine the effect they have on the protein stability. In this project, you will gain knowledge of basic protein formulation, effect from various types processing and excipients and you will get experience with techniques to study protein stability and structure.

  **Supervisor: Lene Jørgensen**

- **Master project in industry or abroad**
  
  You could also do your project in the pharmaceutical industry, Medico industry or at a University abroad (e.g. University of Otago, School of Pharmacy, Dunedin, New Zealand). This requires planning as short notice project are often difficult. You will be asked to supply a copy of your grades, as you will need to work independently. The projects topics in industry are typically defined in detail 2-3 months before the actual start of the project.

  **Supervisors: Lene Jørgensen**
Natalja Genina
Assistant Professor

Research focus: 2D printing/inkjet printing, 3D printing/additive manufacturing, personalized medicine, physicochemical characterization of innovative product, innovative manufacturing, spectroscopy

Information-rich pharmaceutical products

Future pharmaceutical products require new features: we build in personalized elements and include information into the product itself. These products are easily personalized for an individual dose and there is a more straightforward solution for combining several compounds into a single product. Our approach is based on liquid handling processes, such as inkjet printing. Manufacturing of oral dosage forms containing information rich 2D patterns that a smartphone equipped with a suitable app could recognize, could be a way to personalize the drug product. This project will explore the use of printed QR code patterns as a final medicinal product.

Supervisors: Natalja Genina, Jukka Rantanen, Johan Bøtker

Feasibility of printed medicine

Printing technologies enable production of personalized medicine with flexible doses and tailored-release profile. In August 2015, the Food and Drug Administration (FDA) approved the first 3D printed drug. If the needs of the patients are identified, the situation can be different in the near future with more printed drugs coming to the market to provide more exact pharmaceutical treatment in society.

The aim of the project is to 3D print innovative prototype products with the varying geometry, colour, taste, disintegration and dissolution behaviour. The overall goal is to explore the patients’ attitude towards the idea of getting personalized product by conducting interviews and showing the possibilities of 3D printing technology.

Supervisors: Natalja Genina and Sofia Kälvemark Sporrong

Development of personalized medicine by 2D and 3D printing
Delivering an accurate dose of active pharmaceutical ingredients (API) and tailoring their release profile are the key factors to produce efficient pharmacotherapy. The project aims to develop API-containing inks (liquid and/or solid) with suitable characteristics for printing such as viscosity (rheometer), surface tension (pedant drop method) and solubility of API (UV/VIS spectrophotometer, HPLC or thermal analysis). The influence of printing parameters on the stability of the ink and printability will be studied as well. The work will be also focused on preparation (fused deposition modelling and freeze drying) of optimal material for printing by using fused deposition modelling and freeze drying. The overall goal is to produce solid dosage forms with easily adjustable doses and release profiles.

**Supervisors:** Natalja Genina
Lene Jørgensen
Mingshi Yang
Associate Professor

Research focus: Pharmaceutical article engineering, pulmonary drug delivery, and tissue engineering.

Examples of projects:

Spray-dried protein formulations
The spray-drying process may be less expensive, but more efficient to process proteins or peptides as compared to other drying techniques. This project has additional potentials in not only stabilization of the proteins but also engineering the protein formulations into dry particles for various pharmaceutical applications, e.g. inhalable, injectables, and oral administration. This project is intended to investigate the influence of formulation and process variables in the spray-drying process on particle characteristics and physicochemical stability of the protein formulations. This project can be carried out at Novo Nordisk.

Supervisor: Mingshi Yang

Electrospraying/Electrospinning in pharmaceutical application
Electrospraying/Electrospinning is an emerging technology in pharmaceutical field to fabricate micro/nano-structured materials for drug delivery and tissue engineering. This project aim to explore the application of this technology and investigating to promote this technology in pharmaceutical field for producing orodispersible film, wound dressing, and injectable depot microparticles.

Supervisor: Mingshi Yang

Pulmonary drug delivery
Lung diseases including chronic obstructive pulmonary disease (COPD), lower respiratory infections and lung cancer are leading cause of death world-wide. Inhaled drug products can directly reach the sites of the diseases, lower side effect, and improve pharmacological effect. The aim of this project is to utilize particle engineering technology to improve the efficacy of inhaled drug products by improve lung deposition, modulating dissolution rates and absorption of inhaled drugs. This project can be carried out at University of Sydney or Shenyang Pharmaceutical University, China.

Supervisor: Mingshi Yang
Advanced drug delivery by solid lipid particulate systems

Lipid matrix particles can increase the solubility of poorly water-soluble drugs by either increasing the drug solubility or by stabilizing an amorphous or molecular form of drug molecules. The project aims at elucidating the effects of polymorphic forms of lipids on the stability of lipid matrix particles in order to use them as stable carriers in improving the bioavailability of poorly water-soluble drugs for systemic or local therapeutic applications.

Supervisors: Mingshi Yang and Huiling Mu
Anders Østergaard Madsen  
Associate Professor

Research focus: Crystallography, pharmaceutical materials science, synchrotron-based techniques computational pharmaceutics.

Example of projects:

Anhydrate-hydrate systems in pharmaceutical products

The true relationship between the stable anhydrate form and the hemihydrate form is not fully understood, potential leading to issues during the manufacturing of the product as undesired crystal form may suddenly show up. The aim of this project is to understand how the water activity level and the temperature can influence the ratio of the stable anhydrate and the hydrate form of different model compounds.

The relationship will be investigated both using a temperature/humidity X-ray powder diffraction (XRPD) chamber as well as with slurry experiments in different solvent systems (having different water activities) at different temperatures.

Supervisors: Anders Madsen, Jukka Rantanen

Johan Peter Bøtker  
Assistant Professor

Research focus: Computational simulations, image analysis, manufacturing and processing of pharmaceuticals, solid state characterisation, multivariate data analysis.

Example of project:  
Image analysis of pharmaceuticals In this project, image analysis of e.g. the tablet coating quality is investigated using MATLAB programming. The project will revolve around creating MATLAB scripts for segmenting objects in images and constructing and operating analytical scripts as well as artificial neural networks for classification purposes. This project will thus focus on optimizing both the process of obtaining the images and the programming needed to analyse the images.

Supervisors: Johan Peter Bøtker
Jukka Rantanen
Professor

**Research focus:** Process analytical technologies, pharmaceutical materials science, bulk powder analysis, spectroscopy, computational pharmaceutics

**Example of projects:**

Particle properties determine the successful manufacturing of solid dosage forms. Shape and size distributions are critical parameters affecting flow ability of materials. However, it is a huge scientific challenge to translate these particle characteristics into optimal final formulation. There are several new innovative approaches to investigate powder rheology together with solid form analysis, making a huge impact in commercial scale process and product optimization. This project will investigate powder rheology as a part of solid dosage form development, as well as implementation of process analytical technologies (PAT) in an industrial setting. The projects topics in industry are typically defined in detail 2-3 months before the actual start of the project. This project can also be performed as an industrial project at AstraZeneca, Lundbeck or Novo Nordisk.

**Internal supervisors:** Jukka Rantanen
**External supervisors:** AstraZeneca, Lundbeck or Novo Nordisk
Social and Clinical Pharmacy

Social pharmacy seeks to have the broad view of medicines in society and focuses on how medicines are perceived, used, and managed by different actors in society. Clinical pharmacy seeks to ensure the optimal and rational use of medicines for the benefit of patients and society through collaboration between pharmacists, other health professionals, and the patient. SCP’s research ultimately seeks to ensure the benefit of medicines for users, organizations, and society. SCP’s research is concerned with the three levels (the user, organization and society) and how they interact to ensure the optimal use of medicines.

Master thesis projects may be connected to existing research projects in the SCP Group, but other relevant ideas for master thesis projects within social and clinical pharmacy are also welcome.

Areas and ongoing projects are for example:

- Antibiotics use and misuse with focus on low and middle-income countries: user and professional perspectives
- Medicine use among children and young people (study drugs, teaching about medicines among elementary and high schools students, enhancement of self-management of mental disorders)
- Patient safety (medicines at transition from hospital to home, medication-adjustment beds in psychiatric hospitals, hospital-based drug information, pharmaceutical product changes in hospitals)
- The extended pharmacist role (clinical interventions to improve medicines use in hospitals, primary care or across health care sectors, prescribing pharmacists, point-of-care testing, teaching in the local community)
- Pharmaceutical innovation (printing of pharmaceuticals, biosimilarity, genetic-testing technologies) and their impact on society from a regulatory, clinical, and societal view
- New healthcare models including pharmaceuticals: the case of value-based health care and its implementation in the Nordic context. What are the (potential) consequences for patients?
- Developing and testing new community-based qualitative research/advocacy methods for patient empowerment in relation to medicines
- Public discourse on medicines: how are medicines talked about in conventional and social media?
- Development of new communication forms in the community pharmacies
- Patient perspectives on medicines (for example in metabolic diseases, chronic patients accepting the medication/compliance dialogue services, etc.)
- Pharmaco-epidemiology (phase IV-studies, pharmacovigilance, side-effects, register-based studies; efficacy vs. effectiveness, drug utilization)
Supervisors in SCP are: Prof. Anna Birna Almarsdöttir, Assoc. Prof. Lourdes Cantarero-Arévalo, Assoc. Prof. Sofia Kälvemark Sporrong, Assoc. Prof. Lotte Stig Nørgaard, Assoc. Prof. Susanne Kaae, Assoc. Prof. Charlotte Vermehren.

We have a coordinated master thesis student system in the SCP Group. First we accept students for the entire group and then we allocate a supervisor according to competencies needed for the specific master thesis project. If you are interested in learning more about writing a master thesis in SCP, you can meet us at the ‘Master thesis day’ at 26th of October. If you want to write your thesis within SCP, you need to write: lotte.norgaard@sund.ku.dk or anna.birna@sund.ku.dk before November 1st, including the topic you would in interested to write about. The selected students will be notified by November the 9th.
Copenhagen Centre for Regulatory Science

Medical research is a highly multidisciplinary field, which is impacted by scientific advances, new technologies, personalised medicine, and an improved understanding of patient requirements. However, the pharmaceutical development of innovative therapeutic solutions will not advance at the pace consonant with its promise without a simultaneous advance in the development of a flexible regulatory framework. This framework must be agile in meeting the competing goals of fostering innovation and protecting the public health, while integrating approaches to manage scientific uncertainty—the tools and trademark of regulatory science.

**Drug Regulatory Science ecosystem**

With our research, we want to improve the drug regulatory system, by systematically studying its structure and behaviour as well as designing new tools to facilitate regulatory decision-making. Through improvement of the drug regulatory system, we aim to contribute to an improvement of the health of the society. Furthermore, we carry out research that produces evidence to be used in drug regulatory decision-making.

**Marieke De Bruin, CORS Director, Professor in Regulatory Science**

Marie Louise (Marieke) De Bruin was trained as a pharmacist and epidemiologist and has combined academic research with working for the regulatory authorities. She is an affiliate at the Utrecht University, WHO collaborating centre for pharmaceutical policy and regulation in the Netherlands. Furthermore, she was appointed by the European Commission as an independent scientific expert of the Pharmacovigilance Risk Assessment Committee (PRAC 2012-2018), that meets monthly at the European Medicines Agency in London; she is on the steering group of the European Network of Centers for Pharmacoepidemiology and Pharmacovigilance (ENCePP 2017-2019); and she is associate editor of the journal ‘Pharmacoepidemiology & Drug Safety’. Her research focusses on developing new tools, standards and approaches to evaluate the efficacy, safety, quality and performance of medical products in order to assess benefit-risk and facilitate a sound and transparent regulatory decision making.
Christine E. Hallgreen, Assistant Professor

Christine Erikstrup Hallgreen is an assistant professor at the Copenhagen Centre for Regulatory Science. She has a background in engineering physics, and a PhD from Department of Physics at the Technical University of Denmark. In her professional career, she has been previously employed in the pharmaceutical industry (Novo Nordisk) and academia (Imperial College London). Christine has also conducted research at the National Institute of Health, Maryland, USA. In her research, she utilizes her quantitative and methodological training in the development and evaluation of drug regulatory tools and systems. Her research is motivated by a desire to describe and understand the functioning of the regulatory system and thereby optimize and improve the systems and regulatory tools set in place to promote public health. Her research includes development and evaluation of formal qualitative and quantitative methods to assess benefit-risk of pharmaceutical products, probabilistic methods to assess the effect of uncertainty in outcome data, and methods to collect preference values to support benefit-risk decisions.

Current research projects at CORS

How best to protect public health: a comparative analysis of regulatory safety warnings on medicines in Australia, Canada the European Union and the United States.

Medicines have important health benefits but can also lead to harm. When new safety concerns arise, national regulatory agencies issue warnings to health professionals and the public. These warnings differ between countries, but no research has compared the effectiveness of different approaches. This study compares safety advisories on medicines in Australia, Canada, the United States, and the European Union in order to identify how to best protect public health. Within the project several working groups will focus on different aspects: discordances in safety communication, regulatory policy analysis, qualitative analyses on HCP and patient perspectives, media analyses and pharmacoepidemiology to estimate health effects.

Factors ensuring effective Direct to Healthcare Professional Communication of Risk Minimization

This project aims to identify and understand the factors that determine the effectiveness of drug safety communication such as Direct to Healthcare Professional Communication and educational materials and sets out to present suggestions on how to improve the existing communication process.

Similar but not identical: the changing regulatory landscape of biosimilars from the regulator, industry, professional, and patient perspectives

The project aims to explore how different stakeholders interpret “similar,” and how these views impact the approval, development, and use of biosimilars.
The Impact of Paediatric Regulations on Drug Development in a Trans-Atlantic Perspective

The objective of this PhD project is to provide insights into the effectiveness of the EU and US paediatric regulations to promote access to medicines for children as well identifying potential barriers of the paediatric regulations to innovative drug development in general. Ultimately, these analyses should serve to provide recommendations for improvements to the global regulatory frameworks for paediatric medicines development with a focus on the US and the EU.
LEO Foundation Center for Cutaneous Drug Delivery

The aim of the center is to contribute to an integrating knowledge on skin as a barrier for drug absorption, and key properties of drugs and excipients for cutaneous drug delivery. Based on a predominantly physicochemical approach, we aim to develop methods for the rational development of novel drug delivery systems for cutaneous and transcutaneous delivery of both small and large molecules, ranging from wounds to intact skin.

See website for LEO Foundation Center for Cutaneous Drug Delivery

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Novel Analytical Tools

Project examples

Novel Analytical Tools

In vitro modelling of the skin barrier for cutaneous drug delivery

The stratum corneum, the outermost layer of the skin, is the main barrier between the body and its external environment protecting against water loss, and incoming pathogens and/or allergens. It is formed of a complex mixture of dead cells (corneocytes) imbedded in a lipid matrix. Drugs applied topically must pass through this barrier to reach the target; therefore, modelling human skin allows us to predict and understand the ADME properties of topical drugs and design more efficient transport vehicles. Furthermore, skin diseases such as atopic dermatitis and psoriasis are known to affect the lipids present in the stratum corneum, mak-
ing the barrier leakier and the patient more prone to infection. By understanding drug interactions with both healthy and diseased stratum corneum we can better design drugs and formulations to cross the stratum corneum. In this project, you will be isolating stratum corneum from pig ears, and testing the barrier properties of whole stratum corneum and extracted stratum corneum lipids with and without corneocytes. In this line of research, we use physical chemistry and surface science methods to investigate the molecular structure and interactions between the stratum corneum, drugs and formulations. Techniques such as ATR-FTIR, SAXS and neutron reflection will be coupled with more traditional pharmaceutical transport experiments using Franz cells. Using these techniques, we are able to study skin interactions with a wide range of molecules of interest, for example, common topical drugs, dermal penetration enhancers, excipients, formulations, skin irritants and allergens. This project provides an excellent opportunity for you to take control of your studies and steer the project in the direction you find to be the most interesting. It may also be possible to be involved in a neutron reflection experiment at a large-scale neutron scattering facility in the UK or France. The project is expected to be part of one or several publications.

**Supervisor Kathryn Browning**

**Number of Students:** 1

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**Insights into the formation of fibs**

Elastic fibers are important protein-based structures in the extracellular matrix of vertebrates. They provide elastic properties to organs such as arteries, skin and lung and are crucial for their long-term function. A multistep process termed elastogenesis leads to the formation of elastic fibers. It involves a large number of biomolecules and is associated with secretion, deposition and cross-linking of the protein tropoelastin on a scaffold of microfibrils. Although many molecules have been identified to be essential for elastogenesis, their precise interactions are still poorly defined. Investigating the mechanisms of elastogenesis helps not only to understand the functions of matrix components, but also fosters the design of elastin-based biomaterials for tissue engineering, which require biocompatibility and elasticity. This project focuses on investigating interactions of tropoelastin with other matrix proteins during elastogenesis to understand the process of cross-linking and identify binding sites between the cross-linking partners. In particular, we will study interactions between tropoelastin and truncated forms of tropoelastin with enzymatic cross-linkers such as lysyl oxidase-like enzymes and chemical cross-linkers. A range of analytical techniques, including surface plasmon resonance spectroscopy, and solid phase binding assays and quartz crystal microbalance with dissipation monitoring will be used. The results of the study represent a step towards the development of artificial elastic fibers in biomaterials used for tissue engineering and wound healing.

**Supervisor:** Andrea Heinz

**Number of students:** 1.
Development of peptide-based electrospun wound dressings
Wound healing is a major burden to healthcare systems worldwide, and there is a clinical need for dressings that can be used to treat partial thickness burn wounds that affect the epidermal and dermal layers of the skin. Requirements for ideal wound dressings include the ability to prevent infection and maintain skin hydration. The wound dressing should further be non-toxic, non-immunogenic and should enhance tissue regeneration. Peptide-based electrospun wound dressings fulfill these criteria.
This project focuses on the development of an electrospinning method to produce peptide-based wound dressings. The peptide will either be electrospun in a mixture with a polymer or co-axially electrospun (peptide core surrounded by polymer mantle). The electrospun fibers will then be comprehensively characterized by a range of analytical techniques, including scanning electron microscopy, X-ray powder diffraction and differential scanning calorimetry. The release of the peptide from the fibers will also be determined.
Supervisor: Andrea Heinz
Number of students: 1.

Development of peptide-based nano- and microgels for wound healing
Wound healing is a major burden to healthcare systems worldwide, and there is a clinical need for dressings that can be used to treat partial thickness burn wounds that affect the epidermal and dermal layers of the skin. Requirements for ideal wound dressings include the ability to prevent infection and maintain skin hydration. The wound dressing should further be non-toxic, non-immunogenic and should enhance tissue regeneration. Physical and cross-linked peptide hydrogels fulfill these criteria and peptides are interesting building blocks for hydrogels due to their structural role in all biological systems. In physical hydrogels, non-covalent interactions between the self-assembling peptides lead to the formation of an ordered scaffold in water, while cross-linked hydrogels are formed by an appropriate cross-linker. This project focuses on the development of physical and cross-linked nano- and microgels from penta- and hexapeptides. Cross-linking will be achieved through the formation of disulfide bonds or the application of UV radiation to peptides with sensitive functional groups. Moreover, physical hydrogels will be formed from peptides that spontaneously self-assemble to form scaffolds in water. The gels will be characterized in terms of gelation concentration, their structure and size (field emission scanning electron microscopy, circular dichroism spectroscopy, dynamic light scattering) as well as temperature-dependent structural changes.
Supervisor: Andrea Heinz
Number of students: 1.

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**Supervisor:** Andrea Heinz  
**Number of students:** 1

**Novel Biological Models**
There are great and unmet medical needs within the areas of dermatology and venereology. Many skin diseases are characterised by aberrant inflammation and reflect a complex pathogenesis, involving compromised skin barrier function, innate and adaptive immunity, and the presence of microbes. Currently, there are still no good methods for diagnosis and prognosis of disease progression and healing, and subsequently proper disease intervention or treatment of these patients is lacking.

Within this area, we focus on developing novel diagnostics and models for drug delivery in wounds and atopic dermatitis skin, thereby combining drug formulation with techniques related to molecular biology, microbiology, immunology and mass spectrometry.

**Peptide-based drug delivery systems for atopic dermatitis skin infections**
Atopic dermatitis (AD) is a multifactorial relapsing inflammatory skin disease affecting 1 out of 5 Danish children. As decreased levels of antimicrobial peptides are a popular explanation for the observed increased susceptibility to infection in these patients, antimicrobial and immunosuppressing peptides have great therapeutic potential in AD. The goal of this project is the development of potent and safe peptide delivery systems for treatment of AD lesions. The work will comprise optimisation of drug formulation, in vitro antimicrobial and biofilm assays and, if time allows, in vitro immunological assays as well. As we have various peptides to be tested, this project can be performed independently by multiple students.

**Supervisors:** Mariena van der Plas & Martin Malmsten  
**No of students:** 1-3
3D skin models for atopic dermatitis
The goal of this project is to develop 3D skin infection models for studying drug delivery systems in atopic dermatitis. The work will consist mostly of techniques within the fields of immunology and microbiology, including cell and bacterial cultures, analysis of inflammatory markers and signalling pathways, cytotoxicity, and histology.
Supervisor: Mariena van der Plas
No of students: 1

Novel biomarkers for wound infections
Wound healing is a fundamental survival mechanism, and dysfunctions cause significant disease, such as seen in infections after burns, trauma and surgery, as well as in non-healing ulcers. Currently, the prevalence of non-healing wounds is estimated to be over 40 million worldwide, a number projected to rise with 6-9% annually, due to aging of the population and the increasing incidence of diseases that contribute to non-healing ulcer development, such as obesity and diabetes.
Project 1 The goal of the first project is to investigate the function of an abundant antimicrobial protein, highly expressed in diabetic ulcers but not in venous leg ulcers, and whether it can be used as a biomarker and/or treatment for certain types of wound infections. The work will consist of in vitro techniques within the fields of molecular biology, microbiology and immunology.
Project 2 The goal of the second project is to investigate the effect of bacterial enzymes on protein cleavage patterns and peptide generation in wounds. The work will consist of in vitro molecular biology and microbiology techniques, as well as mass spectrometry (under supervision) and subsequent data analysis.
Supervisors: Mariena van der Plas & Jun Cai
No of students: 1 per project

Development of novel peptide-based antimicrobial dressings for wound healing
With the increasing occurrence of resistance in bacteria, development of alternatives to antibiotics are essential. The goal of this project is the development of antimicrobial and immunomodulatory peptide-based dressings using electrospinning. The work will comprise optimisation of peptide-incorporation in/on the electrospun fibres, analysis of peptide-release kinetics, in vitro antimicrobial assays and, if time allows, in vitro biofilm and immunological assays as well.
Supervisors: Mariena van der Plas & Andrea Heinz
No of students: 1

Development of smart wound dressings for sensing of infections
The goal of this project is the development of smart electrospun dressings containing sensors that reporting changes in e.g. temperature, pH, inflammation and bacterial contamination. The work will comprise development and optimisation of sensor incorporation in electrospun
fibres and testing of the generated dressings using a range of techniques within microbiology, immunology and molecular biology.

Supervisors: Mariena van der Plas & Andrea Heinz
No of students: 1

Master projects in Lund (Sweden)
Would you like to do your Master project abroad, but without having to move? We have close collaborations with excellent groups at Lund University, with interesting projects in the fields of host-pathogen interactions and structural protein biology in relation to wound and skin. Contact us for more information.

Supervisors: Mariena van der Plas and local supervisor