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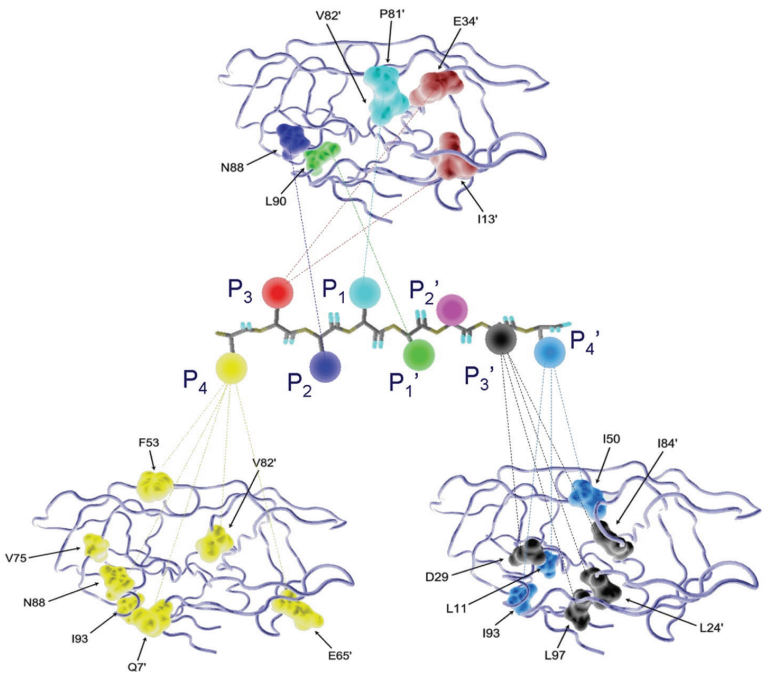
ANNUAL REPORT 2007

Department of Pharmaceutical Biosciences



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ANNUAL REPORT 2007

Department of Pharmaceutical Biosciences

Editing committee: Eva Brittebo, Marianne Danersund, Björn Hellman and Mats Karlsson
Layout: Magnus Jansson
Front page illustration: Aleksejs Kontijevskis
"Cross-dependencies of amino acids in retroviral proteases and their substrates required for high catalytic activity as determined by proteochemometric modeling"

Introduction

This report gives a short overview of the research projects and some of the events that occurred during 2007 at the variety of divisions and disciplines that make up the Department of Pharmaceutical Biosciences. We have experienced a number of audits and quality assessments of our research as well as our undergraduate and graduate programmes. The evaluation of research at Uppsala University, called Quality and Renewal 2007, was carried out during the spring. The international panel together with the chairman Alexander T Florence from University of London met department members for interviews, presentations, and laboratory visits. The panel concurred that "pharmaceutical informatics and neuropharmacology of drug addiction are two areas of vital importance for the future of the department. Pharmacometrics is of high relevance to the pharmaceutical industry, in the development of new drugs and for optimising therapies of marketed drugs. Proteochemometrics is an emerging methodology to assess interactions between proteins and small molecules as in receptor-ligand interactions. Research activities in neuroscience and drug dependence are well integrated and focused on the comprehension of drug addiction mechanisms. The drug safety research has a high potential with regard to successful development of in vitro test systems for early identification of teratogenic compounds."

Research also underpins all our teaching activities. Without research projects in the areas of drug dependence, pharmacology, pharmaceutical biochemistry, pharmacokinetics, pharmacometrics and toxicology we would not be prepared to meet the needs of the students in pharmacy, biomedicine, medicine and chemical engineering. Many of the pharmacy students perform their degree project at the department but we also encourage them to spend a period of study in other European countries, North America or Asia.

The department has a high priority for research training of our PhD students. In the second half of 2007 the department was getting ready for the peer review of its graduate programmes conducted by the National Agency for Higher Education. This report will be published in the spring 2008. In July ten PhD students participated in the 8th ULLA Summer School in Leiden/Amsterdam, the Netherlands. ULLA is a European Consortium for postgraduate training in the field of pharmaceutical sciences. Pharmacy schools from Uppsala, London, Leiden, Amsterdam, Copenhagen, Paris and Parma are members of the consortium. The ULLA Summer School allows PhD students from all these faculties to widen their knowledge regarding drug discovery, drug development and drug management as well as other issues facing the pharmaceutical companies. Not less important, the Summer School gives PhD students an opportunity to create a European network and to have a great time.

The teachers, laboratory, technical and administrative staff and PhD students are the key resource of the department. Some department members have moved on and others have joined. During 2007 the following new senior academic staff members joined the department: Sven Björkman was appointed Professor in Pharmacokinetics, Malin Andersson was appointed Research fellow in Imaging mass spectrometry for studies of peptides and proteins and Andrew Hooker was appointed Lecturer in Pharmacometrics. We look forward to working with them.

A challenge facing the department relates to the cost of premises as well as operating and maintenance costs for our laboratories that are spread within the large BMC building. To address this issue the department has examined the possibility of relocating some of the laboratories. The next year we will start relocation and

condensation of some divisions. This will provide new opportunities for staff and students and at the same time improve the cost of premises.

During the first half of 2007 Lennart Dencker was holding the position as Head of Department. After many years serving as the Dean of the Faculty and Head of Department Lennart Dencker resigned from these duties in July. He will now focus on research and teaching but he will also continue with special faculty missions. His efforts were essential and much appreciated and we thank him for all dedicated hard work during the years. Now I will carry on and rely heavily on the advice of him and all other members of the department.

Uppsala March 5, 2008
Eva Brittebo

List of Contents

Organization

Chairman
Eva Brittebo (from July 2007)
Lennart Dencker (until June 2007)

Deputy chairman
Mats Karlsson

Department board
Eva Brittebo, chairman
Marianne Danersund, secretary
Mathias Hallberg, teacher representative
Margareta Hammarlund-Udenaes, teacher representative
Mats Karlsson, teacher representative
Anne-Lie Svensson, teacher representative
Björn Hellman, teacher representative
Lennart Dencker, teacher representative, deputy
Maria Norlin, teacher representative, deputy
Raili Engdahl, technical/administrative representative
Agneta Hortlund, technical/administrative representative
Marina Rönngren, technical/administrative representative, deputy
Mats Nilsson, graduate student representative
Angelica Quartino, graduate student representative
Sadia Oreland, graduate student representative, deputy
Rasmus Sjölin, student representative (from July, 2007)
Ludvig Möller, student representative, deputy (from October, 2007)

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Lennart Paalzow

Director of graduate studies
Ernst Oliw

Directors of undergraduate studies
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Annika Häger
Uwe Rossbach
Student representative (vacant)

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Kjell Berggren
Raili Engdahl
Ronnie Hansson
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Lena Norgren

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Ronnie Hansson
Björn Hellman
Annika Hipeli
Andrew Hooker
Emma Lundkvist
Anders Nyholm
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Rikard Sandström
Anne-Lie Svensson
Maria Swartling
Erik Wågstrand

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Marianne Danersund
Agneta Hortlund
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Karin Svensson
Gunnel Wiberg
Kjell Åkerlund

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Raili Engdahl
Britt Jansson
Birgit Jansson
Britt-Marie Johansson
Angela Lannerbro
Lena Norgren
Jessica Strömgren

Scientific Reports

Division of Biological Research on Drug Dependence

Biological Research on Drug Dependence

Fred Nyberg and Mathias Hallberg

(I) Studies on neuropeptides, neurohormones and steroids in relation to opioid sensitivity and chronic pain (including animal experimental models, *in vitro* cell cultures and clinical studies). In clinical studies the project directs to assessment of genetic polymorphism in various transmitter systems combined with quantification of neuropeptide levels in various body fluids. Peptides chosen for analysis includes the tachykinins and opioid peptides. In animal studies models for nociceptive, neuropathic pain are chosen.

(II) Studies on neuropeptides, neurohormones and steroids (in particular anabolic androgenic steroids = AAS) in relation to drug dependence (including experimental animal models, *in vitro* cell cultures and clinical studies). In clinical studies the project directs to assessment of genetic polymorphism in various transmitter systems combined with quantification of neuropeptide levels in various body fluids. Peptides chosen for analysis includes the tachykinins and opioid peptides. In animal studies models to investigate opiate tolerance and withdrawal and drug self-administration (in collaboration with other laboratories) are used. Endogenous peptides with high potency to attenuate withdrawal reactions have been identified and serve as basis for design and synthesis of peptides and non-peptides that may be further developed to act as drugs in the treatment of opiate addiction. In studies of effects of AAS on the brain neurochemical technologies (radioimmunoassays, autoradiograhy, Western blot, etc.) are combined with various behavioral assays.

(III) Studies on the functions of growth hormone (GH) and prolactin (PRL) and their receptors in the central nervous system (including experimental animal models, in vitro cell cultures and clinical studies). Receptors for GH have been identified in the brain in areas of relevance for many of the known effects of GH on the central nervous system (CNS). Beneficial effects of GH on cognitive functions are recorded by the assessment of memory and cognition using the Water maze in conjunction with various neurobiological techniques.

(IV) Studies on atypical opioid peptides (endomorphins, hemorphins and casomorphins) in relation to behaviour and mechanisms for their release.

(V) Studies on synthetic compounds acting on angiotensin receptors. Receptor assays specific for the AT1, AT2 and AT4 receptors are used to guide synthesis and design of peptide and non-peptide analogues. Compunds with high affinity and selectivity are further studied with regard to agonist activity in functional assay *in vitro* or *in vivo*.

Members of the group during 2007

Fred Nyberg, Professor
Mathias Hallberg, PhD Associate Professor
Qin Zhou, Farm Dr. Researcher
Kristina Magnusson, PhD Student
Tobias Johansson, PhD Student
Milad Botros, PhD Student
Uwe Rossbach, PhD Student
Martin Elfverson, PhD Student
Dan Henrohn, PhD Student
Jenny Johansson, PhD Student
Britt-Marie Johansson, Technician
Alfhild Grönbladh, Research Associate

Publications 2005-2007

1. Kastrup Y, Le Greves M, Nyberg F and Blomqvist A (2005) Distribution of growth hormone receptor mRNA in the brain stem, and spinal cord of the rat. *Neurosci 130, 419-425.*
2. Hallberg M, Kindlundh AMS and Nyberg F (2005) The impact of chronic nandrolone decanoate administration on the NK1 receptor density in rat brain as determined by autoradiography. *Peptides 26, 1228-1234.*
3. Steensland P, Hallberg M, Kindlundh AMS, Fahlke C, Nyberg F (2005) Amphetamine-induced aggression is enhanced in rats pre-treated with the anabolic androgenic steroid nandrolone decanoate. *Steroids, 70, 199-204.*
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Reviews

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Growth hormone, insulin and insulin-like growth factor-1: Do they interact at the blood-brain barrrier.; in Nyberg F (ed.): The somatotrophic axis in brain function. *Elsevier Academic Press, San Diego, 2006, pp 75-79.*
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Neuropeptides in hyperthermia. *Progr Brain Res 162, 2007; 162:277-293.*

Dissertations 2007

1. Milad Botros, Farm Lic
Characterization of Specific Binding Sites for Substance P (1-7) in the Rat Central Nervous System

Agencies that support the work/Funding 2007

Swedish Research Council Medicin 450.000 SEK/year, total 1 350 000 SEK
Precision Science System, 2 100 000 SEK
Swedish Foundation for Strategic Research, 200 000 SEK
Berzelii Centre for Biotechnological Research, 600 000 SEK
Disciplinary Domain of Medicine and Pharmacy. 400 000 SEK
Swedish National Drug Policy Coordinator, 500 000 SEK

Division of Medical Mass Spectrometry

Medical Mass Spectrometry

Malin Andersson

My main project utilizes MALDI imaging mass spectrometry (IMS) for the topographical elucidation of proteins, neuropeptides and neurotransmittors and their changing concentrations in the brain during physiological and pathophysiological events. As large genomic and proteomic datasets are being generated from homogenates of various tissues and pathological diseases, the need for information on the spatial localization of transcripts and their encoded protein products has become more pressing. The structural heterogeneity and complexity of many tissues, such as brain, requires an approach that provides high anatomical resolution coupled with quantitative analysis of proteins in the context of unbiased assessment. Ultimately, visualizing molecular expression in tissue sections in 2 and 3-dimensional space using MALDI IMS will provide insights into the neural substrates of neurodegenerative disease pathogenesis and help pave the road toward improved and tailored treatments.

Members of the group during 2007

Malin Andersson, Research Assistant
Maria Fälth, PhD Student
Anna Nilsson, PhD Student

Publications 2005-2007

1. Groseclose, R. M., Andersson, M., Hardesty, W. M., and Caprioli, R. M. (2007).
   Identification of Proteins Directly from Tissue: In Situ Tryptic Digestions Coupled with Imaging Mass Spectrometry. *Journal of Mass Spectrometry 42 (2):254-262.*
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   Microdialysis Coupled with Liquid Chromatography / Mass Spectrometry; Handbook of Microdialysis: Methods, Applications and Perspectives, *Elsevier Science. 16: 251-266*

Agencies that support the work/Funding 2007

Swedish Research Council 2 681 000 SEK
Disciplinary Domain of Medicine and Pharmacy 279 000 SEK

Division of Pharmaceutical Biochemistry

Stress Adaption

Matti Lang

Cells adapt to stress by modulating the expression of genes which are part of their defense and adaptation machinery.

We are investigating the molecular mechanisms of how stress, in the form of toxic chemicals, oxidative stress and experimental psychologicals stress modulate gene expression.

Stress responding genes used as models include;

Cyp2a5 /Cyp2a6; The encoded enzymes are involved in detoxification of xenobiotics. The genes were chosen because they are inducible by a variety of toxic chemicals and also by viral and bacterial infection and by oxidative stress. In addition, conditions which disturb transcriptional activity lead to sustained high expression of these genes via mRNA stabilization.

iNos; the enzyme: inducible nitric oxide syntase protects organisms against microorganisms and is upregulated by oxidative stress under infestation.

P53; the protein is a transcription factor playing a central role in control of cell growth. The gene is upregulated for example by DNA damage caused by xenobiotics.

Cyp2B1, CYP2E1, CYP2D1 ; genes encoding for drug metabolising enzymes are used as models to see how psychological disorders such as psychosis and depression influence their level of expression and the level xenobiotic metabolism, and thereby affect patients sensitivity to drugs.

A central part of our research strategy and goals is to seek and identify stress response elements (stress sensors) on the mRNA and DNA of these model genes, responsible for their regulation under stress. And to identify transacting factors interacting with these elements. Our working hypotheis is that genes responding similarly to stress should have similar stress sensors and transacting, stress activated, factors.

Current status;

We have identified hnRNPA1 (heterogenous nuclear ribonucleoprotein A1) as a key regulator of the *Cyp2a5 and Cyp2a6*. The protein is activated by different toxic insults and can regulate the Cyp2a5 expression both at transcriptional and posttranscriptional levels: by interacting with the promoter alternatively with the 3´-UTR of the corresponding mRNA. As the hnRNPA1 is multifunctional, it ispossible that it controls the gene expression at different levels on the gene expression pathway (for refs., see below).

Two other members of the hnRNP family; the hnRNPI and hnRNPL, were found to interact with the iNOS mRNA in an infection dependent manner, and with a possible regulatory function (see ref, below). We are currently working on the detailed mechanisms of gene regulation by these proteins.

We have identified hnRNPc as an important regulator of P53 vie a regulatory element in the first exxon of the encoding mRNA. HnRNPc is activated by DNA

damage and disturbed trancription threby linking the toxic reaction to upregulation of P53.

A strong downregulation of CYP2E1 and CYP2B1 has bee shown in relation to psychosis, and evidence has been obtained for the involvement of the dopaminergic signalling pathway in this process.

Members of the group during 2007

Matti Lang, Professor
Kyle Christian, PhD student

Publications 2005-2007

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Steroid P450

Kjell Wikvall and Maria Norlin

Our research is focused on the properties and regulation of cytochrome P450-mediated enzymatic processes involving steroids.

1) Bioactivation and metabolism of vitamin D and cholesterol (Principal investigator: Kjell Wikvall)
2) Steroids in hormonal signalling (Principal investigator: Maria Norlin)

The Steroid P450 group is part of UBAP (Uppsala Bioactivation Program), a scientific program for bioactivation (metabolic activation) at the Faculty of Pharmacy.

1) Bioactivation and metabolism of vitamin D and cholesterol (Principal investigator: Kjell Wikvall)

This research is focused on enzymes and genes of importance for vitamin D bioactivation and cholesterol homeostasis. Effects on these processes by endogenous and pharmacological compounds are studied. Vitamin D is needed for regulation of calcium levels in the body and vitamin D deficiency leads to skeletal diseases such as rickets in children and osteomalacia/osteoporosis in adults. The biologically active form, 1α,25-dihydroxyvitamin D3, is formed through metabolic activation. The activated form of vitamin D blocks cell division and increase cell differentiation. Vitamin D analogues are used in the treatment of psoriasis and are of potential interest in cancer treatment. For these reasons, it is important to obtain more knowledge about the enzymes that activate and metabolize vitamin D and the roles of the formed metabolites.

Excess cholesterol and disturbances in cholesterol balance may lead to health problems such as heart disease, gall stones and neurological disease. Cholesterol is eliminated from the body by being converted into bile acids. Bile acids and a type of cholesterol derivatives called oxysterols have recently been shown to be ligands to nuclear receptors which regulate genes in lipid homeostasis and drug metabolism. It is therefore important to obtain more information about the enzymes that form and metabolize bile acids and oxysterols, such as their properties and how they are regulated. During the last couple of years these processes have gained increasing interest in connection with development of new drugs to treat abnormal cholesterol levels.

An ongoing project concerns a new group of side-chain modified 15-oxosterols, synthetic inhibitors of cholesterol biosynthesis, which are potential drugs for treatment of high cholesterol levels. Other studies involve influences of anti-epileptic and anti-viral drugs on the processes described above, which result in adverse side-effects in some patients.

2) Steroids in hormonal signalling (Principal investigator: Maria Norlin)

This research concerns steroids involved in hormonal signalling in connection with sex hormone biosynthesis, neurosteroid function and cellular proliferation. The studies are focused on physiological and pharmacological control of steroid levels, effects of metabolic events and mechanisms for regulation of gene expression. Some of the steroids of interest in this area are dehydroepiandrosterone (DHEA) and 5α-androstane-3β,17β-diol.

DHEA is well-known as a precursor for androgens and estrogens but also plays roles in brain function and in connection with cell growth and functions of

the immune system. This steroid has been proposed as a potential drug for treatment of several diseases, for instance systemic lupus erythematosus, an autoimmune disease. 5α-androstane-3β,17β-diol, an estrogenic hormone, is believed to play a role for hormone-dependent proliferation, particularly in the prostate.

Several enzymatic reactions of interest, involving the steroids mentioned above, are catalyzed by CYP7B1, a multifunctional enzyme which impacts the levels of a number of steroids in many different tissues. For instance, CYP7B1 is responsible for enzymatic conversions that affect prostate hormone levels and the concentration of neuroactive steroids in the brain. This enzyme may be a potential future target for therapy aimed at regulating the levels of steroids of importance for abnormal cell growth, immune function or in neurodegenerative processes.

Current studies involve regulation of the levels of DHEA and other neurosteroids in neurons and glial cells and mechanisms for estrogen receptor-mediated control of the CYP7B1 gene by endogenous steroids and pharmaceutical compounds.

Members of the group during 2007

Kjell Wikvall, MD, PhD, Professor
Maria Norlin, PhD, Associate Professor
Maria Ellfolk, PhD Student
Johan Lundqvist, PhD Student
Hanna Pettersson, PhD Student
Wanjin Tang, PhD Student

Publications 2005-2007

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   Hormonal regulation of the human CYP27A1 and CYP7B1 genes
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Agencies that support the work/Funding 2007

The Swedish Research Council-Medicine: 350 000 SEK (Kjell Wikvall)
Åke Wiberg´s Foundation: 100 000 SEK (Maria Norlin)

Division of Pharmacology

Proteochemometrics

Jarl Wikberg

Research during 2007 was concentrated on 1) the continued development of the **Bioclipse** platform and 2) the further development of proteochemometrics.

During the year Bioclipse 1.2.0 was released (available at www.bioclipse.net). Among its many previous features Bioclipse now also contains: 1) An interactive console view for writing scripts in Javascript, 2) extension point for descriptor calculations from chemical structures, i.e. mixing different descriptor providers, 3) ability to create CDK descriptors and JoeLib descriptors using a wizard, 4) a matrix editor for editing spreadsheet-like files with custom matrix implementations (Jama is the default implementation), 5) Chart view for displaying charts from e.g. the matrix editor, 6) experimental support for BioMoby, and much more.

During the year many proteochemometrics studies directed to validation of the technology in drug design, drug monitoring and protein engineering were completed as follows: 1) A proteochemometric study on the four serotypes of dengue NS3 proteases was completed, 2) studies demonstrating how one can use proteochemometrics in design of improved antigens for antibodies was completed, 3) a study how to apply proteochemometrics to predict inhibition by novel and previously unseen compounds for all common CYP forms was completed, and 4) a study using clinical susceptibility data for HIV protease inhibitors for prediction of resistance to clinically used HIV retardants from HIV genome sequence was developed (available at www.hivdrc.org). A prediction server based on Bioclipse software for prediction of drug resistance based on HIV protease sequence was set up and is available at www.hivdrc.org.

In ongoing studies assay systems for a multitude of resistance mutated HIV proteases was also set up and a large number of organic compounds were synthesized based under experimental design and proteochemometric design principles with the aim to develop new HIV protease inhibitors with a broad spectrum over a large array of resistant HIV forms.

During the year Bioclipse won yet another international price, the Trophées du libre 2007 special price of the Jury (see http://www.tropheesdulibre.org/Bioclipse.html).

Development of Bioclipse and validation of proteochemometrics continues.

Members of the group during 2007

Jarl Wikberg, Professor
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Ola Spjuth, PhD Student
Martin Eklund, PhD Student
Eskil Andersen, Programmer
Jonathan Alvarsson, Programmer

Carl Mäsäk, Programmer
Bjarni Juliusson, Programmer
Ramona Petrovska, Technician

Publications 2005-2007

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Agencies that support the work/Funding 2007

The Swedish Research Council, 500 000 SEK
The Swedish International Cooperation Agency, 600 000 SEK
The Swedish Research Links, 200 000 SEK
The Swedish Institute, 370 000 SEK

Prostaglandin

Ernst H. Oliw

Arachidonic acid and a few other polyunsaturated fatty acids are bioactivated in humans by enzymatic oxygenation to prostaglandins, leukotrienes, epoxides (EETs) and other local hormones, which contribute to fever, pain, inflammation and cancer development, and to regulation of physiological processes during reproduction and in many other organs. Common drugs such as aspirin, acetaminophen (paracetamol) and ibuprofen inhibit biosynthesis of prostaglandins and reduce symptoms of disease, but may also cause side effects related to their actions. Other drugs are based leukotriene receptor antagonists (e.g., montelukast), which are use for treatment of bronchial asthma. Bioactivation of polyunsaturated fatty acids also occur in plants and fungi where oxygenation of linoleic and linolenic acids is important for the plant-pathogen interaction and for fungal reproduction and pathogenicity. The goal of our research is to investigate the mechanism of oxygenation and bioactivation of fatty acids and to determine their biological function.

We investigate mainly three groups of enzymes: (i) lipoxygenases, (ii) cytochromes P450 and (iii) heme-containing dioxygenases. These enzymes occur in man but also in important fungal pathogens, e.g., *Aspergillus fumigatus* causing farmer´s lung disease and *Magnaporthe grisea*, causing rice blast disease and destruction of 25% of the rice crop of Japan. Our goal is to understand how the enzymes work in order to understand their physiological and pathophysiological functions and to develop new drugs.

In humans, the prostaglandin endoperoxide, PGH_2, can be transformed by cytochromes P450 to thromboxanes, prostacyclin and to 19-hydroxy-PGH_2, the precursor of 19-hydroxy-PGE_2. The latter is the main prostaglandin of human seminal fluid and occurs in high concentration in human semen, where it is formed by CYP4F8 of the seminal vesicles. CYP4F8 and CYP4F22 are also expressed in skin and we investigate their oxygenation of fatty acids.

All lipoxygenases contain a catalytic metal, iron in humans and plants. We focus our basic research on the first described manganese-lipoxygenases, which are important for Gäumannomyces graminis, an important pathogen of wheat, and its structurally similar lipoxygenases of *Magnaporthe grisea*, and *Aspergillus fumigatus*. These fungi also contain oxygenate cyclooxygenase-related enzymes, which oxidized linoleic acid by to a series of vicinal diols (5,8-dihydroxy-, 7,8-dihydroxy- and 8,11-dihydroxyoctadecadienoic acids) via formation of hydroperoxides (8-hydroperoxy- and 10-hydroperoxylinoleic acid), which likely function as sporulation hormones. The reaction mechanism and identification of these diol synthases are described in our recent paper in *The Journal of Biological Chemistry* (Ref. 14).

Members of the group during 2007

Ernst H. Oliw, MD PhD, Professor
Ulrike Garscha, PhD student
Tomas Nilsson, PhD student
Fredrik Jernerén, PhD student

Publications 2005-2007

1. Cristea, M., Engström, Å., Su, C., Hörnsten, L., and H. Oliw , E.H.(2005) Expression of manganese lipoxygenase in *Pichia pastoris* and site-directed mutagenesis of putative manganese ligands, *Arch. Biochem. Biophys. 434, 201-211.*
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Agencies that support the work/Funding 2007

The Swedish Research Council Medicine, 300 000 SEK
The Swedish Research Council Formas, 459 000 SEK

Novel transformations of polyunsaturated fatty acids and eicosanoids.

Ernst Oliw, Johan Bylund and Tomas Nilsson

Arachidonic acid can be oxygenated to biologically important mediators of fever, pain and inflammation , viz. prostaglandins, leukotrienes and epoxyeicosatrienoic acids (EETs). We focus on the oxygenation of arachidonic acid and eicosanoids by cytochrome P450 4 family enzymes: CYP4F8 (prostaglandin H 19-hydroxylase) and two orphan enzymes, CYP4F22 and CYP4V2. Mutations of two latter have been implicated in retinal and skin diseases and we are now expressing these enzymes in yeasts in order to characterize these enzymes.

Characterization of heme-containing fatty acid dioxygenases and hydroperoxide isomerases of human and plant pathogens

Ernst Oliw, Ulrike Garscha, Fredrik Jernerén

Fungi are severe pathogens of man and can be devastating for important crops. Aspergillus causes farmer´s lung disease and invasive aspergillosis of immunocompromized patients. Rice blast disease is caused by Magnaporthe grisea, and destroys ~25% of rice crops worldwide. Aspergillus and M. grisea contain cyclooxygenase-related enzymes, diol synthases and dioxygenases, which transform linoleic acid into hydroperoxides and dihydroxy fatty acids. Our aim is to characterize the enzymes by enzyme expression, gene targeting and by studies on their biological importance.

Drug Dependence

Ingrid Nylander

The projects within the research group focus on the neurobiological substrates for individual differences in vulnerability for alcohol dependence, with special emphasis on the impact of early environmental factors. Alcohol dependence is a complex trait and the phenotype related to vulnerability for dependence is based on the interaction of multiple genes and environmental factors. Adverse experiences during the critical childhood and adolescence periods can cause long-term neurobiological and behavioural alterations and increased vulnerability for psychopathology, including drug dependence. The environment may also provide protection, for instance in a predisposed individual. The mechanisms underlying the environmental influence are not fully understood. The projects within the research group aim to elucidate mechanisms underlying protective and risk factors for excessive alcohol consumption. For that purpose animal experimental models are used in combination with extensive evaluation of neurobiological and behavioural consequences of different early environmental conditions.

A rodent maternal separation (MS) model is used to simulate different environmental settings. Rat pups are separated from the caregiver short (15 min, MS15) or prolonged (360 min, MS360) periods during the first postnatal weeks. MS15 is more similar to natural conditions where the mother regularly leaves the litter for shorter periods of time. Previous results within the group provide evidence for MS15 being protective. Adult rats subjected to MS15 during the first weeks of life have a low alcohol intake. In addition, genetically predisposed rats subjected to MS15 exhibit a slower acquisition of alcohol intake. The prolonged separations interfere with early social interactions and are used to simulate an emotional stressful environment for the rat pups and/or the mother. MS360 is associated with an increased risk for excessive alcohol intake. Rats subjected to MS360 have higher alcohol consumption, they prefer higher alcohol concentrations and in alcohol-preferring rats, MS360 rearing adds to the risk as evidenced by an even higher adult alcohol intake. Some rats do not respond to the emotional stressful early environment and the reason for these differences in responsiveness are to date not known. In projects within the group, possible brain target systems mediating the early environmental influence are studied. Focus is on neuropeptides, such as opioids, oxytocin and vasopressin, and monoamines that are important for early social behaviour and normal neuronal development. It is hypothesized that disruption of early developmental processes in these transmitter networks cause long-term changes in behaviour and, in turn, alcohol consumption later in life.

Members of the group during 2007

Ingrid Nylander, Professor
Erika Roman, PhD, Post-Doc to July 2007
Chris Pickering, PhD, Post-Doc from July 2007
Stefan Schlussmann, PhD, Guest Researcher
Sadia Oreland, PhD Student
Lisa Gustafsson, PhD Student (examined 2007)
Marita Berg, Technician

Publications 2005-2007

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Reviews 2005-2007

1. Roman E, Nylander I.
   Review. The impact of emotional stress early in life on adult voluntary ethanol intake – results of maternal separation. *Stress 8 (2005) 157-174.*

Dissertations 2007

1. Gustafsson Lisa
   Endogenous Opioids and Voluntary Ethanol Drinking. A comprehensive evaluation of the impact of the early-life environment in rats.
   Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Pharmacy 51
   http://publications.uu.se/abstract.xsql?dbid=7776

Agencies that support the work/Funding 2007

The Swedish Research Council (K2005-04X-12588-08A) 200 000 SEK
The Alcohol Research Council of the Swedish Alcohol Retailing Monopoly (98/21) 130 000 SEK
AFA Insurance, 500 000 SEK

Neuropeptides

Lisa Gustafsson, Sadia Oreland

Endogenous opioid peptides are sensitive to early environmental factors as evidenced by specific short- and long-term MS-induced effects. Rats reared in a stressful environment have signs of a dysfunctional opioid system. They have characteristics similar to alcohol-preferring rats, lower basal opioid levels and an enhanced response to alcohol. A more detailed analysis of effects on opioids in specific brain regions, opioid receptors and studies of treatment outcome with opioid receptor antagonists in rats reared in different environments are subject for examination. MS-induced effects on oxytocin and vasopressin are currently studied.

Monoamines

Sadia Oreland

Recent results provide evidence for MS-induced effects on gene expression of 5-HT receptors. In particular, specific alterations have been shown in rats reared in the protective environment, MS15, as compared to other rats and these results may give further insight in protective mechanisms. Ongoing studies examine alcohol-induced effects on monoamines in animals reared in different environmental settings.

Neuronal Development

Chris Pickering

Previous studies have found an affect of early life stress on development of the nervous system. We have observed decreases in expression of hippocampal NMDA and AMPA receptors which suggests dramatic changes in glutamate, the excitatory neurotransmitter in the brain. Therefore, we are currently investigating developmental effects both during and immediately following MS. By measuring changes in neuron or glia number and several markers of synapse formation, we can describe the mechanism of MS and how these changes can lead to the differences in alcohol consumption that we observe in older animals.

Neuropharmacology

Lena Bergström

Reports from police, Customs, and medical services all point to the fact that abuse of anabolic androgenic steroids (AAS) in the society is increasing. The abuse leads to personality changes often of violent nature. There are investigations showing that misuse of AAS often is associated with an increased abuse of other addictive drugs including alcohol, amphetamine and opiates. There is a very limited knowledge how AAS affects the brain and the aim of this project is to study the effects of AAS on the reward system in the brain and in regions associated with aggressive behaviour. The reward system i.e. neuronal circuits which are activated following a positive stimuli (including drugs of abuse) are principally very similar in all mammals. The studies are therefore performed on laboratory rats long-term treated with AAS in different doses. We have started studies in order to investigate whether long-term treatment with AAS will change the activity in the reward system and done some interesting observations. In one study dopamine and its metabolites, HVA and DOPAC, were measured in the extracellular fluid in the nucleus accumbens using a microdialysis technique. We found reduced levels of the dopamine metabolites which we interpreted as a reduced dopaminergic activity, and the observation might explain why abusers of AAS also consume other illegal drugs in a higher extent. The results are followed up with measurements of mRNA for monoaminergic enzymes, transporters and receptors in order to conclude the mechanisms behind the decreased dopaminergic activity. Like humans, rodents show an increased aggressive behaviour when injected with AAS and there is known that certain brain regions in the brain (amygdala, hypothalamus, PAG) are activated during an aggressive attack. Aggressive behaviour increases when the level of the neurotransmittor serotonin is decreased or when serotonin receptors are lacking. On the contrary drugs that increase the serotonin activity will decrease aggression. GABA and glutamate are important transmitters in the these neuronal circuits having inhibitory and excitatory functions, respectively. We are measuring markers for these transmitter systems using quantitative real time PCR in purpose to find specific changes.

Members of the group during 2007

Lena Bergström, Associate Professor
Carolina Birgner, PhD student

Publications 2005-2007

1. Birgner C., Kindlundh-Högberg A., Ploj K., Lindblom J., Nyberg F., and Bergström L.
   Effects on rat brain dopamine and DOPAC levels after sub-chronic nandrolone administration followed by an amphetamine challenge., *Pharmacologyonline, 2007*
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Neurogenesis and Neurodegeneration

Anne-Lie Svensson

The research focus on the role of neurosteroids on neurogenesis and of interactive processes that is ongoing in neurodegenerative disorders like Alzheimer disease, with emphasis on neuroprotective properties of neurosteroids against amyloid-β-induced toxicity. Neurosteroids are produced in brain in the presence of steroidogenic enzymes. Specific neurosteroids are endogenous modulators of neuronal functions responsible for many biological and pathophysiological effects. Some neurosteroids might play important roles in cognitive functions. Normal aging is associated with several alterations in neurosteroid production and secretion. Decreases in their levels might contribute to aging of the brain and to loss of important nervous functions, such as memory. However, the mechanisms of their action at cellular and molecular level are not well understood.

A plausible link between neurosteroids and neurodegenerative disorders like Alzheimer´s disease (AD) has been discussed. AD is characterized pathologically by deposits of amyloid plaques in the cortex and hippocampus of the brain. The principal component of amyloid plaques is the amyloid-β peptide. Amyloid-β peptide is known to play a central role in the pathogenesis of AD, through the ability of amyloid-β monomers to aggregate and form protofibrils. Amyloid-β has been implicated in cell death during the course of AD and exerts toxic effects on neurons both *in vivo* and *in vitro*. Identification of compounds able to prevent Aβ formation, aggregation and thereby prevent protofibril formation is an important goal of the therapeutic strategies of AD.

The significance of neurosteroidogenesis in regulating neurodegenerative mechanisms is unknown. Accumulation of amyloid-β, incuced by toxic events in cells, might be able to reduce the synthesis of neuroprotective neuroteroids, thus favour/support neurodegenerative processes. The aim of this research work is to further and deeper study neuroprotective properties of neurosteroids and their metabolites, against amyloid-β-induced toxicity, as well as the underlying molecular mechanism(s), with focus on neurogenesis and apoptosis. Brain reductions of neurosteroids might be relevant to the pathophysiology and therapeutics of AD.

Members of the group during 2007

Anne-Lie Svensson, Associate Professor
Marie Eketjäll, PhD Student
Elise Persson, Research Assistant

Publications 2005-2007

1. A Östergren, A-L. Svensson, N. L. Lindquist and E.B. Brittebo
Dopamine melanin-loaded PC12 cells: a model for studies on pigmented neurons. *Pigment Cell Res, 18 (2005) 306-314*

Behaviour

Erika Roman

The concept of ethoexperimental studies of behaviour has been launched to promote the advantage of integrating ethology and experimental psychology. With this approach, the aim is to use test conditions and procedures that are based on the circumstances and challenges the animal meets under natural conditions. A laboratory for behavioural tests in rodents has been established and is under continuous development. The laboratory comprises ethologically founded tests, including tests for assessment of neonatal development, exploratory behaviour, locomotor activity, anxiety-like behaviour, learning and memory and a multivariate test arena (the multivariate concentric square field, MCSF) and utilizes multivariate statistical approaches. The MCSF test is designed to include opportunity for exploration, risk assessment, risk taking, shelter seeking and approach and avoidance behaviour in rodents. The guiding principle of the MCSF test is that it is unprejudiced, i.e. the test is not designed to measure a particular mental condition. Instead the test situation involves a free choice of different environmental settings and items that provide the opportunity to assess essential features of the animal's mentality. In this way a behavioural profile is generated. The projects involve development and validation of the MCSF test in order for the test to be used in studies including motivation for seeking rewards, learning and memory mechanisms and repeated trial effects. Research in the field of e.g. drug dependence and eating disorders has focused mainly on the consumption and less attention is paid to the actual motivation or drive behind these behaviours. By inserting stimuli possessing rewarding properties in a device attached to the side of the risk area in the MCSF, it is possible to evaluate how prone the animals are to take the risk of crossing the risk area in order to reach the reward. Ongoing studies assess the animal's motivation for passing the risk area by increasing the resistance of passing. The association between natural rewards, such as sexual activity and food intake, and drugs of abuse, i.e. ethanol, is subject for examination. Furthermore, selectively bred ethanol-preferring and ethanol-avoiding rats are examined with regard to their behavioural profiles and aspects of motivational and consummatory behaviours (collaboration with Dr. Petri Hyytiä, Finland; Professors Giancarlo Colombo, Italy and Lawrence Lumeng, USA). Besides these projects, advice is given in projects related to behavioural neuroscience and collaborations have been established with the Departments of Pharmaceutical Biosciences and Neuroscience, UU.

Members of the group during 2007

Erika Roman, Ph.D.
Bengt J Meyerson, Professor Emeritus

Publications 2005-2007

1. Roman E, Gustafsson L, Hyytiä P, Nylander I.
   Short and prolonged periods of maternal separation and voluntary ethanol intake in male and female ethanol-preferring AA and ethanol-avoiding ANA rats. *Alcoholism: Clinical and Experimental Research (2005), 29, 591-601.*

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3. Meyerson BJ, Augustsson H, Berg M, Roman E, Nylander I.
The Concentric Square Field: a multivariate test arena for ethoexperimental analysis of explorative strategies, in Proceedings of Measuring Behavior 2005, 5th International Conference on Methods and Techniques in Behavioral Research, LPJJ Noldus, F Grieco, LWS Loijens, PH Zimmerman (Eds). *Noldus Information Technology, The Netherlands, 2005, pp. 459-460.*
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The Concentric Square Field: a multivariate test arena for analysis of explorative strategies. *Behavioural Brain Research (2006), 168, 100-113.*
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The multivariate concentric square field test reveals different behavioural profiles in male AA and ANA rats with regard to risk taking and environmental reactivity. *Behavioural Brain Research (2007), 183, 195-205.*

Reviews 2005-2007

1. Roman E, Nylander I. The impact of emotional stress early in life on adult voluntary ethanol intake – results of maternal separation in rats. *Stress (2005), 8, 157-174.*

Agencies that support the work/Funding 2007

The Swedish Brain Foundation, post-doc stipend, 2007-2008, 250 000 SEK
Disciplinary Domain of Medicine and Pharmacy, 150 000 SEK
The Alcohol Research Council of the Swedish Alcohol Retailing Monopoly (07-21:1), 130 000 SEK

Division of Pharmacokinetics and Drug Therapy

Pharmacokinetics/Pharmacodynamics

Margareta Hammarlund-Udenaes and Sven Björkman

Our research aims to improve the understanding of drug distribution and elimination in relation to drug effects. In particular, this includes experimental and clinical studies of CNS active drugs and their transport to the brain. Pharmacokinetic and pharmacodynamic principles are also applied to the clinical use of drugs, in order to design rational dosage regimens.

Members of the group during 2007

Margareta Hammarlund-Udenaes, Professor
Sven Björkman, Professor
Malin Alenius, PhD Student
Jörgen Bengtsson, PhD Student
Emma Boström, PhD Student
Markus Fridén, PhD Student
Ulrika Gillespie, PhD Student
Stina Syvänen, PhD Student

Publications 2005-2007

1. Bengtsson, J., B. Jansson, and M. Hammarlund-Udenaes
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The use of a deuterated calibrator for in vivo recovery estimations in microdialysis studies. *J Pharm Sci, 2007.*
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32. Hammarlund-Udenaes, M., M. Friden, S. Syvanen, and A. Gupta
On The Rate and Extent of Drug Delivery to the Brain. *Pharm Res, 2007.*
33. Lundquist, P., P. Hartvig, G. Blomquist, M. Hammarlund-Udenaes, and B. Langstrom
5-Hydroxy-L-[beta-11C]tryptophan versus alpha-[11C]methyl-L-tryptophan for positron emission tomography imaging of serotonin synthesis capacity in the rhesus monkey brain. *J Cereb Blood Flow Metab, 2007. 27(4): p. 821-30.*
34. Lundquist, P., M. Roman, S. Syvanen, P. Hartvig, G. Blomquist, M. Hammarlund-Udenaes, and B. Langstrom
Effect on [11C]DASB binding after tranylcypromine-induced increase in serotonin concentration: positron emission tomography studies in monkeys and rats. *Synapse, 2007. 61(6): p. 440-9.*
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Synthesis of two potential NK1-receptor ligands using [1-11C]ethyl iodide and [1-11C]propyl iodide and initial PET-imaging. *BMC Med Imaging, 2007. 7: p. 6.*

Reviews 2005-2007

1. Björkman, S.
A comment on the application of drug tissue-plasma partition coefficients K(p) in eliminating organs to calculation of volume of distribution at steady state. *J Pharmacokinet Pharmacodyn, 2005. 32(5-6): p. 655-8.*
2. Björkman, S.
Prediction of cytochrome p450-mediated hepatic drug clearance in neonates, infants and children : how accurate are available scaling methods? *Clin Pharmacokinet, 2006. 45(1): p. 1-11.*

Dissertations 2007

1. Emma Boström
Pharmacokinetics and Pharmacodynamics of Oxycodone and Morphine with Emphasis on Blood-Brain Barrier Transport
Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Pharmacy 50
http://publications.uu.se/abstract.xsql?dbid=7772

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Swedish Academy of Pharmaceutical Sciences, 317 000 SEK

Blood-brain barrier transport of drugs – mechanisms and methods

Margareta Hammarlund-Udenaes, Jörgen Bengtsson, Markus Fridén, Stina Syvänen, Ulf Bredberg (AstraZeneca), Mats Bergström (GSK), Bengt Långström (Uppsala Imanet), Yoshiharu Deguchi (Japan), Tetsuya Terasaki (Japan)

Our research is focused on understanding how the blood-brain barrier (BBB) functions regarding drug transport in health and disease, and to optimise methods to measure brain penetration of drugs. The research is important for the drug industry that has problems in finding good drug candidates for brain diseases, partly due to a lack of understanding of which parameters to optimise for.

One of our purposes is to find the key parameters that describe BBB transport of drugs. This has been accomplished during 2007 after 15 years of work within the area, and is summarized in a publication in Pharmaceutical Research, "On the rate and extent of drug transport to the brain" (e-pub ahead of print in 2007). Another important publication during 2007 was the paper by Fridén et al (Drug Metab Dispos), where two in vitro methods were compared with microdialysis to describe intra-brain distribution of drugs, this with the purpose of finding optimal and rapid methods in early drug discovery to better choose between drug candidates regarding blood-brain barrier transport. The paper has already obtained international attention. Positron Emission Tomography (PET) is also used as a way of understanding BBB transport of drugs. Among other work, species comparisons are made in order to understand the value of using rodents to extrapolate to the human situation. Oxycodone was specifically studied and Emma Boström defended her thesis during 2007. Her main finding was that the opioid drug oxycodone is actively taken up at the BBB, resulting in 3 times higher unbound concentrations in the brain than in blood. The transporter responsible for this uptake is investigated in research collaboration with researchers in Japan. When identified and connected to the properties of the drug substances, it could be used to optimise active uptake of drugs into the brain.

Clinical Pharmacy Research

Margareta Hammarlund-Udenaes, Malin Alenius, Ulrika Gillespie, Håkan Melhus (Clinical Pharmacology, Uppsala), Claes Mörlin (Medicine, Uppsala), Åsa Kettis-Lindblad (Social Pharmacy, Uppsala), Per Hartvig (Univ of Copenhagen) and Leif Lindström (Uppsala)

This research is divided into two parts, the first being a development of a method to describe drug effects in a naturalistic setting of psychosis patients. This is an area where clinical drug development today uses selected subpopulations to measure new drug effects. The research is also aiming at finding correlations between genetic factors, and other treatment factors between responders and non-responders, and between those without and with side effects, in order to optimise drug treatment to this patient group.

In the other part, we are interested in measuring the results of clinical pharmacist interventions in acute medical care, with a specific focus on readmissions of patient 80 years and older. The purpose of this research is to see if and if so, how the contributions of clinical pharmacy services in the hospital ward changes patient

treatment and status. This research area is new in Sweden and important for the development of this area of work for pharmacists.

Clinical pharmacokinetics of coagulation factors VIII and IX

Sven Björkman, with professor Erik Berntorp, M.D. and research associate Karin Lindvall, R.N., Malmö.

The aim of the project is to optimize the prophylactic treatment of haemophilia with coagulation factors VIII and IX by the use of individually tailored dosing. Prophylactic treatment of severe haemophilia with coagulation factor concentrates is effective but very expensive, with a cost approaching or even exceeding SEK 1 million per patient per year. Optimizing the dosing of factor VIII or factor IX by means of clinical pharmacokinetic (PK) principles can potentially yield important benefits both from a purely medical as well as from an economical point of view. The project started in 1989 and has resulted in widespread international acceptance of "pharmacokinetic dosing" in this particular field of disease management. The activity during 2007 comprised:

- Examination of the disposition of factor VIII as a function of age of the patient by means of population PK modelling. It is especially urgent to fill the gaps in our knowledge on the disposition of this factor in pre-school children since prophylactic treatment of haemophilia normally starts during the second or third year of life.

- Testing the feasibility to achieve dose-tailored haemophilia prophylaxis by daily self-injection of factor concentrate instead of by the conventional every two days or three times per week treatment schedules.

- Evaluating the PK and clinical information obtained during extensive licensing studies on a novel factor VIII preparation, Advate (Baxter Inc.).

Pharmacometrics

Mats Karlsson

Pharmacometric research focuses on nonlinear mixed effects ("population") models. Such models describe data, generally is the response-time profiles observed in a clinical trial, by a basic model, accounting for the general structure of the underlying system, a a set of hierarchical variability components, accounting for variability between subjects, within subjects over time and remaining between observation variability. Research at the pharmacometrics group can be divided into four main areas. First, development and evaluation of methods for efficient and robust model building. This involves development of estimation algorithms, methods for model diagnosis and sequential procedures for model building. The result of the research, when applicable, is made available as free software. Secondly, so-called platform models are being developed for the use in specific therapeutic areas or for particular therapeutic/pharmacological principles. Such a model may involve the time-course a system biomarker or a set of such biomarkers during normal, diseased or provoked situations. The third research area concerns utilization of the developed models for the purpose of designing studies, deciding upon dosing strategies and other developmental decisions. Last, we also do analyses of dose-concentration-response data from trials to understand therapies with existing drugs with the aim of allowing improved therapy.

Members of the group during 2007

Mats O Karlsson, Professor
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Andrew Hooker, Senior Lecturer
Rikard Sandström, Senior Lecturer
Ulrika Simonsson, Senior Lecturer
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Niclas Jonsson, Adjunct Professor
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Joy Dansirikul, Post-doctoral Fellow
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Matts Kågedal, PhD Student
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Petra Jauslin, PhD Student
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Publications 2005-2007

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Dissertations 2007

1. Mats Magnusson
Pharmacodynamics of Enzyme Induction and its Consequences for Substrate Elimination
Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Pharmacy 52
http://publications.uu.se/abstract.xsql?dbid=7812
2. Jakob Ribbing
Covariate Model Building in Nonlinear Mixed Effects Models
Digital Comprehensive Summaries of Uppsala Dissertations from the Faculty of Pharmacy 59
http://publications.uu.se/abstract.xsql?dbid=7923

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Clinical modelling of pharmacokinetics in HIV, TB and malaria therapy

Doaa Elsherbiny, Mats Karlsson, Ulrika Simonsson

Plasmodium falciparum, the human immunodeficiency virus (HIV), and Mycobacterium tuberculosis are three devastating pathogens in tropical areas. Due to the geographical overlap of malaria, HIV and TB prevalence, the diseases are likely to co-exist in a great number of individuals. For these individuals, there is an obvious need for concomitant use of antimalarial, antiretroviral and antitubercular drugs. Drug-drug interactions may result from concurrent administration of drugs leading to diminished therapeutic efficacy of or increased toxicity from one or more of the administered drugs. Drug-drug interactions are an important concern in the management of patients with HIV because of the large number of antiretroviral drugs and other drugs that are required by these patients for the management of co-morbidities and opportunistic infections. Combination therapy has also been introduced in the management of malaria and TB, to overcome drug resistance. Limited information is available in the literature on drug-drug interactions

between the artemisinin antimalarial drugs and other drugs such as antiretrovirals or antitubercular drugs; consequently, the extent of such interactions is not fully known.

Activities of CYP enzymes and consequently drug-drug interactions occurring due to their inhibition or induction can be studied by using probe drugs. The pharmacokinetics of probes and drugs under investigation can be described by mathematical models in order to characterise and quantify the interaction. We have developed enzyme turnover models to describe the time course of induction of different CYP450 enzymes by different artemisinin derivatives. One focus of the work has been to compare the potential for drug-drug interactions among the artemisinin drugs to choose a derivative that is suitable for combination therapy from a drug-drug interaction perspective.

One commonly used antitubercular drug is rifampicin which is know to induce CYP450 enzymes. Our work has involved quantitative analysis by modelling the pharmacokinetics of other drugs metabolised by these enzymes and which are used in HIV treatment. For example, the population pharmacokinetics of nevirapine in HIV-infected patients taking nevirapine-based antiretroviral therapy in the presence and absence of the antitubercular drug rifampicin has been evaluated.

Pharmacokinetic drug-drug interactions can possibly be compensated for by dose adjustment of the target drug. The developed nevirapine model was used for simulations of different doses of nevirapine which revealed that increasing the dose of nevirapine to 300 mg twice daily elevated nevirapine concentrations above subtherapeutic levels in most patients, with minimum exposure above the recommended maximum concentration. We have also investigated the population pharmacokinetics of lopinavir in TB/HIV co-infected children taking lopinavir/ritonavir in a ratio of 1:1 in the presence of the antitubercular drug rifampicin, with that of lopinavir in HIV-infected children taking lopinavir/ritonavir in a ratio of 4:1. Increasing the ritonavir dose in the TB/HIV co-infected children resulted in model predicted lopinavir trough concentrations above the recommended minimum therapeutic concentration.

Development of glucose-insulin models for Type II diabetes

Joy Dansirikul, Bengt Hamrén, Petra Jauslin, Mats Karlsson, Jakob Ribbing, Hanna Silber, Ulrika Simonsson

To characterize the functionality of the glucose insulin system in Type II diabetic mellitus (T2DM) patients and healthy volunteers a multitude of clinical trial types are used. Experimental provocation studies include: clamping of glucose or insulin by variable rate infusions, intravenous bolus administration of glucose or insulin, administration of oral glucose solution or administration of test meals with well-characterised nutrient content. The studies vary in length from a few hours to about a day. In each subject the time-profile of glucose and insulin is measured. We have developed an integrated mathematical model that based on simultaneous analysis of both glucose and insulin time-profiles in all subjects can quantitatively describe the result of such experiments. This model, which includes production, disposition and control (homeostatic) mechanisms have shown to be able to realistically simulate the outcome of trial studies at the raw data level.

Medium-term clinical trials in T2DM patients, varying in length from a few weeks to about a year usually focus on measured longitudinal changes in fasting plasma glucose, fasting insulin and the fraction glucosylated hemoglobin (HbA1c). As hemoglobin in red blood cells has a life-span in the body of a few months and the glucosylation of hemoglobin is a reaction directly dependent on the concentration of glucose HbA1c is a suitable marker of long-term glycemic control. It is elevated

in T2DM patients. Based on data from large-scale clinical trials in T2DM patients and non-diabetic subjects with hypertriglyceridaemia and abdominal obesity we have developed a mathematical model that quantifies the link between plasma glucose concentration and HbA1c. This model is based on mechanistic aspects of the production and elimination of red blood cells and hemoglobin as well as relationships between fasting glucose and daily average glucose. In a complementary model the relationship between insulin sensitivity, glucose production and disposition and changes in beta-cell mass has been charaterised and quantified in the same populations. Both models can realistically simulate the outcome of clinical trials with respect to glucose, insulin and HbA1c.

The models have been developed for the purpose of being able to quantitate changes in the system following interventions (drug administration, diet changes) and associate these with known or hypothesized mechanisms of impact of the system. Further the models are intended as tools for hypothesis generation regarding single or combined interventions as well as clinical trial design optimization.

Dose individualisation in paediatric transplantation

Lena Friberg, Mats Karlsson, Johan Wallin

Cyclosporin and tacrolimus are two commonly used drugs in pediatric transplantation. For the last 20 years, virtually all renally transplanted children in Finland have been monitored for their plasma drug concentrations by the Clinical Pharmacology group at the University Hospital in Helsinki resulting in a unique data base. In collaboration with this group, and including also other therapy information from these patients, we are analyzing the data with the following aims: (i) to optimize a pre-transplantation test procedure with respect to convenience and information content, (ii) to characterize determinants of variability in pharmacokinetics for this population over time after transplantation, and (iii) to outline the relationship between plasma drug concentration and biomarkers/ clinical endpoints in order to allow better decision criteria for dose adjustments. A similar project involving the drug tacrolimus is underway in collaboration with hospitals in Stockholm, Gothenburg and Brisbane.

Mechanism-based pharmacokinetic models

Mats Karlsson, Grant Langdon, Rocio Lledo, Mats Magnusson, Rikard Sandström, Lotfi Slimani

Clinical pharmacokinetic experiments typically measures drug concentrations in plasma only. As a consequence, pharmacokinetic models typically used in drug development aim to describe, with the minimum model complexity, these observations of drug concentration in plasma. Such models have limited capacity to predict concentration-time profiles in tissues and organ. Also, mechanistic insight about drug disposition dependence of factors related to individual organs and tissues may not be possible to incorporate in a fully satisfactory manner. Physiologically-based pharmacokinetic (PBPK) models, which have a structure based on anatomy, can provide predictions in tissues and organs. However, because of their complexity, such models are not used for analyzing clinical data. We have showed that for a relatively simple PBPK model such analyses can become feasible by using informative prior information about physiology and drug-related parameters. To further improve such an approach we are combining information about (co-)variability in organ and tissue properties from a data-base representing physiological values for about 30000 subjects, tissue composition models and models for relating drug molecular properties and in vitro data to expected

behaviour in tissues and organs.

The impact of induction properties for a drug candidate in drug development or for a drug already in clinical use can influence the use of the drug itself and have major impact on the metabolism and transport of other drugs used in combination with the inducing drug. Prediction of the time course and extent of induction is complex. It depends on the halflife of the induced enzyme(s) and transporters, the pharmacokinetics and dosing regime of the inducing agent, and the relationship between the plasma concentration of the inducer and extent of the induction. If it is to be possible to predict the activity of enzymes and transporters at any point in time during and after the induction, all of these aspects have to be understood. By developing mechanistic models, the key elements of these systems can be isolated, and their contribution to the induction process can be determined. This is done by using methods to assess the activity of the enzymes and transporters in vivo by using different probes that have specific reactions for certain enzyme such as midazolam (CYP3A4), coffeine (CYP1A2) or that are transported by a specific transport protein such as digoxin (Pgp). In vitro data from liver microsomes can also successfully be incorporated into the models to explain and predict the kinetics of the drug and the risk of potential drug-drug interactions.

Oncology

Lena Friberg, Emma Hansson, Andrew Hooker, Mats Karlsson, Angelica Quartino, Johan Wallin

Within the oncology area, three PhD-students are working on projects related to extensions and applications of an earlier developed semi-physiological model describing the time-course of myelosuppression that has been successfully applied for numerous anticancer drugs. For docetaxel, the model has during 2007 been optimized to improve simulation properties as shown in predictive checks. In addition, information on both total leucocytes and neutrophils were modeled simultaneously. Using a Sigmoid Emax-model for the drug effect improved the model's ability to more accurately predict the magnitude and time of nadir for docetaxel. To further improve the understanding of the feedback system regulating the feedback and thereby improve the predictive capacity of the model, a clinical study has been initiated in collaboration with the Department of Oncology where endogenous G-CSF concentrations following chemotherapy are measured. Approximately half of the intended number of patients has been included and a preliminary interim analysis has been performed.

We have shown inter-occasion variability in the myelosuppression model parameters to be relatively low compared with inter-individual variability and to be similar in magnitude across 4 data sets including different drugs and drug combinations. This indicates that neutrophil counts may be used for model-based dose-adaptations. In a simulation study we showed the superiority of using neutrophil counts over drug concentrations for dose-individualization. The predictive performance was also good when only a couple of samples per treatment cycles were used to guide dosing optimization. For etoposide an Excel macro has been built for individual dose-reductions and dose-escalations to reach a by the clinician determined nadir based on measured drug concentrations and/or neutrophil counts. In patients who develop febrile neutropenia, we have seen that there is no major difference in the shape of the time-course of myelosuppression compared with other patients who develop Grade 4 neutropenia. Therefore the only option to avoid life-threatening febrile neutropenia may be to avoid Grade 4 neutropenia, for example by predictive covariates or neutrophil-guided dose-adjustments.

The myelosuppression model has also been applied on leukocyte counts in rats. Data were obtained following administration of one of six different anticancer drugs. The drug-related parameter, Slope, was close to the values earlier estimated based on patient data, particularly after correction for species-differences in protein binding (etoposide) and in bone marrow sensitivity shown earlier in the CFU-GM assay (5-fluorouracil). The results indicate that the full time-course of myelosuppression can be predicted for a new drug based on drug-specific parameters estimated from rat data and drug-independent system-related parameters determined in patients.

The group has also been involved in several projects aiming to individualize anticancer drug dosing based on genotype, phenotype and grade of liver-impairment for taxanes and irinotecan.

Antibiotics

Lena Friberg, Mats Karlsson, Elisabet Nielsen

We aim to improve on the understanding of pharmacokinetic-pharmacodynamic relationships for antibiotics of value for improving dosing recommendations and minimizing resistance development. A semi-mechanistic model including one population of growing, susceptible bacteria and one population of resting bacteria has been developed based on constant concentrations of 5 different drugs. In vitro experiments have now been performed where the drug concentrations diminish with half-lives observed in patients and data from these experiments can be reasonably well predicted based on the developed model and its parameters. Similar experiments have been performed for gentamicin. Following modelling, the results will be used in combination with our population pharmacokinetic model developed from data in a prospective study in term and preterm neonates. Based on current guidelines on goals for peak and trough concentrations, preterm neonates need a higher starting dose and longer dosing intervals than the current gentamicin guidelines suggest.

Colistin is an old drug which was abandoned because of its toxicity. With emerging antibiotic resistance the drug is now in use again as a last rescue and has shown to be promising for its synergistic effects. A LC-MS-MS method to quantify colistin and its prodrug CMS in plasma and broth have been developed and applied to analyse colistin concentration in six patients. The half-life of colistin appear to be longer than what has earlier been anticipated based on chemical analysis methods with lower sensitivity and selectivity.

Clinical trial design

Andrew Hooker, Stefanie Hennig, Kristin Karlsson, Mats Karlsson, Rocio Lledo, Joakim Nyberg

There are two principled ways by which models can be used to help optimizing trial designs for information regarding parameter estimates. The first is by simulation from the model and a proposed designed followed by parameter estimation from the resulting data set. The simulation, repeated many times with different random seeds, thus provides measures of precision and bias of parameter estimates. With this methodology we have investigated differences in different randomization schemes for dose-finding trial. It was found that dose-randomized trials are more powerful to characterize the underlying relation. This increase in power can be achieved with in most instances a similar or lower number of observed side-effects. The second way of optimizing trial designs is through formal estimation of design parameters. A number of different criteria can be used to optimize designs. We have developed methods and software (PopED) to do so for ED-optimal designs,

which take into account that the underlying system (model) is not known before the study takes place. While optimal design previously has focused on optimization of sampling times, we have extended this to apply also to other aspects of trial designs, such as the dose administered and the length of run-in, treatment and wash-out phases.

Pharmacodynamic modelling in other disease areas

Lena Friberg, Anders Grahnen, Andrew Hooker, Kristin Karlsson, Mats Karlsson, Maria Kjellsson, Brigitte Lacroix, Anna Lönnebo, Guangli Ma, Elodie Plan

Apart from the disease areas described above we are working on pharmacodynamic models for several other effects and adverse events. All antipsychotic drugs cause prolactin elevations because they compete with dopamine's tonic inhibition of peripheral D2 receptors causing prolactin release. Increased prolactin concentrations can cause unpleasant adverse events. We have developed a mechanism-based agonist-antagonist interaction model describing the time-course of prolactin following antipsychotic drug administration based on both patient and healthy volunteer data. The model include circadian variations and a feedback loop that explains the observed tolerance development due to that dopamine release increase following prolactin elevations and the increased dopamine concentrations reduces the prolactin release caused by the antipsychotic drug. The tolerance development has previously been described by depletion of a prolactin pool and we have now shown that the agonist-antagonist interaction model is superior in describing the time-course of prolactin for different drugs and a range of different of dosing schedules.

A longitudinal transition model describing the probability for 20% improvement in rheumatoid arthritis and the probability for drop-out has been developed where the probabilities for transition depend on covariates, drug concentrations of certolizumab pegol and previous score. The model has successfully been used for simulations to show differences of different dosing strategies. An exposure response model of certolizumab pegol in Crohn's disease has been developed and simulations from the model explained outcomes of different study designs in performed studies and dosing strategies in different subgroups of patients.

A pharmacodynamic model of the effect of budesonide on ACTH and cortisol has been built that can serve as a tool for further understanding of the hypothalamic-pituitary-adrenal (HPA) axis and be useful in the development of drugs interacting with the axis. A production driven by surges was shown to be superior over production driven by a sum of cosine functions.

A joint disease progression model for three stroke scales assessing neurological and/or functional deficit were developed, i.e. for the NIH stroke scale, Scandinavian stroke scale and Barthel index. A joint dropout model was used and correlations between the interindividual variabilities in the three scales were investigated. This kind of model has potential to utilise data across different scales collected in different trials.

The time course of sleep stages has been characterized and its relation to placebo and drug effects using Markov models in patients with insomnia. Good simulation properties of the model were demonstrated and simulations have been performed to investigate the efficacy of different dose levels.

The efficacy of anti-epileptic drugs are measured as number of seizures per day, i.e. count data. A Poisson model for the count data has been developed and the significance of including overdispersion and Markovian elements has been shown.

Model building methodologies and estimation methods

Paul Baverel, Kristin Carlsson, Andrew Hooker, Mats Karlsson, Rada Savic

Pharmacometric models are based on (patho-)physiological and pharmacological knowledge. The complexity and heterogeneity of biological data makes the knowledge about and development of statistical data analysis methods a central part of this scientific field. There are many benefits of using pharmacometric models to analyze data from clinical trials, for example the ability to handle sparse data and to integrate different types of observations in one model. One drawback, on the other hand, is that they are complex and intrinsically non-linear which together with the fact that the amount of data from a single clinical trial can be large (thousands of data points from hundreds of patients with multiple response variables) often make computer run-times non-ignorable. Generally, run-times can be divided into short (minutes), intermediate (hours to days) and long (days to months). The number of runs in a complete analysis tends to range between 30 and many hundreds.

Linear models allow for closed form solutions to aspects such as estimates of parameter precision, and influence of single observations. Non-linear models, on the other hand, generally have no such solutions. At best, there may be numerical approximations (estimates of standard errors is one example) but being approximations, it is usually unknown how good they are in any particular application. Computer intensive and resampling based methods, for example the bootstrap and stepwise covariate model building, are designed to replace complicated closed form solutions or approximations by brute force computerized calculations. This project aims at develop methods for pharmacometric model development and evaluation. One typical example is rational and statistically correct algorithms for adding explanatory variables, .i.e. covariates, to the models. Another example is simulation based criteria for model qualification. One integral part of these research activities is the implementation of the methods in freely available software to facilitate a wider and consistent use of the new algorithms. Examples of software developed by the group are PsN and Xpose.

Division of Toxicology

Embryotoxicology

Lennart Dencker

Embryogenesis The embryo is unprotected from pharmaceuticals and environmental pollutants. The intended pharmacological effects of drugs are often exerted in the conceptus as well. They can be reversible, but have occasionally detrimental morphological downstream effects. We try to understand the molecular background of malformations by monitoring the "symphony" and its different tunes concerted by the collected expression of genes and proteins governing embryonic development and brain maturation. We use this information from embryos, applied to murine and human embryonic stem cells, to develop mechanism-based in vitro test systems to reveal the teratogenic potential of substances. Using the antiepileptic and teratogenic drug valproic acid, an HDAC inhibitor, and some less teratogenic analogues of valproic acid, we try to visualize e.g. by gene ontology studies which categories of genes may be responsible for the teratogenic action (such as neural tube defects). They seem not necessarily to be HDAC-related. Such studies in the embryo and in embryonic stem cells in parallel, combined with the extensive literature on the role of individual genes and pathways in morphogenetic processes, give us new biological information which can be applied in creating tools for screening purposes in drug development and classification of environmental chemicals. In addition, physiological systems such as hemodynamics during development is approached, in this case the widespread property among pharmaceuticals to affect potassium (IKr)-channels thereby affecting rhythm also in the embryonic heart, leading to blood pressure and circulatory fluctuations, and thus oxygen supply, being deleterious to the morphological development of vessels and other embryonic organs. This latter research is lead by adjunct professor Bengt Danielssson.

Neurogenesis Several chemicals exert estrogenicity, being a potential problem esp. during development (reproductive organs and sex-specific neurobehaviour). There is relatively little known about the mechanisms behind their sex specific brain development in general, especially regarding the impact of the sex chromosomes. Chicken and Japanese quail are commonly used models, and we are studying basic sex differences (with and without estrogen exposure) in gene expression in their brain. We have shown that mainly sex chromosome-bound genes are differentially expressed in the male and female embryonic brain very early and steadily thereafter. There were no clear-cut effects of estrogens in gene expression, while there were some sex differences in the fetal peptidome, with a general up-regulation of peptides (about 60 identified) in diencephalons with embryonic age, as well as in the overall effects of EE_2 detected by multivariate analysis, with a potential specific alteration in a HPG-axis related candidate (GnIH-RP2). Many chemicals (incl. drugs) given to newborn mice disrupts normal brain (growth spurt) development, resulting in disturbed spontaneous behavior in adulthood. Polybrominated diphenyl ethers (PBDEs) are environmental

contaminants found in human and animal tissues worldwide. We have investigated their short-term effects on protein expression in hippocampus, striatum and cortex by using two-dimensional difference gel electrophoresis (2D-DIGE). We determined the identity of 111 differentially expressed proteins in cortex, 39 (35%) of which are known to be cytoskeleton-related. As in striatum, we found elevated levels of the neuron growth-associated protein Gap43 in the cortex. Although much work is require to make a complete picture of these early changes, it is a beginning of a mechanistic approach to a potentially important general health problem caused by environmental chemicals as well as drugs.

Members of the group has been involved in projects carried out on the BMMS

Members of the group during 2007

Lennart Dencker, Professor
Bengt R Danielsson, Adjunct Professor
Michael Stigson, Researcher
Camilla Svensson, Researcher
Kim Kultima, Researcher
Henrik Alm, PhD Student
Måns Jergil, PhD Student
Mats Nilsson, PhD Student
Birger Scholz, PhD Student

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Agencies that support the work/Funding 2007

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The Swedish Association of the Pharmaceutical Industry, 270 000 SEK

Bioactivation and Toxicity

Eva Brittebo

Our current studies are directed towards characterization of toxicant-induced biochemical processes leading to cell damage in the nasal mucosa, vascular tissues (endothelial cells) and pigmented neurons. In addition, the delivery of drugs and chemicals to the brain via the nasal olfactory pathways is being examined.

Bioactivation and toxicity of xenobiotics (Anna Franzén, Elena Piras, Helén Andersson)

A high expression of drug-metabolizing cytochrome P450 (CYP) enzymes in specific cells in the upper respiratory tract makes this tissue sensitive to chemicals that are bioactivated. We have identified a number of nasal olfactory toxicants, e.g. dichlobenil, chlorthiamid, 2,6-dichlorophenyl methylsulfone and methimazole. All these compounds are bioactivated by nasal CYP to reactive metabolites that become irreversibly bound to the airway epithelium in vivo. In vitro studies revealed an extensive CYP2A5-mediated bioactivation of dichlobenil and 2,6-dichlorophenyl methylsulfone in olfactory microsomes whereas no activation occurred in liver microsomes. The CYP enzymes reside in the endoplasmic reticulum (ER) and this cell organelle may therefore be a preferential target for reactive metabolites. We observed that upregulation of the ER stress protein GRP78 and activation of the ER resident caspase 12, are early and cell specific events following exposure to nasal olfactory toxicants activated by CYP2A5. The cellular upregulation of GRP78 and activation of caspase 12 as well as the cellular formation of protein adducts in nasal glands colocalize with the early cellular lesions induced by nasal toxicants.

Epidemiological and experimental animal studies suggest that inc reased air pollution contributes to cardiovascular diseases. Humans are exposed to polyaromatic hydrocarbons (PAH) and nitro-PAHs by inhalation of diesel exhausts. We have previously reported that PAHs are bioactivated and induce DNA damage in cultured human endothelial cells (HUVEC) following pretreatment with CYP1A1-inducers. We are currently studying the bioactivation and effects of nitro-PAHs in HUVEC. In addition, the cellular expression of drug metabolizing CYPs in the highly vascularized human endometrium is examined. Our data on the cellular expression and activities of major endometrial CYP forms will be useful when human in vitro test are to be developed for detecting chemicals affecting the embryo implantation process.

Food mutagens and the neurotoxin BMAA (Anna Östergren, Oskar Karlsson and Nils Gunnar Lindquist)

The ß-carbolines norharman and harman are formed at cooking of food. These compounds structurally resemble the Parkinson-inducing toxicant MPTP (1-methyl 4-phenyl 1,2,3,6-tetrahydropyridine), known for its ability to damage neuromelanin-containing dopaminergic neurons of the substantia nigra. The ß-carbolines showed an affinity to melanin and were retained for at least a month in neuromelanin-containing neurons. Norharman was found to induce neurodegeneration, glial cells activation and motor impairment in mice. Furthermore, these compounds induced ER stress and cell death in cultured PC12 cells. An in vitro model of dopamine melanin-loaded cultured PC12 cells was developed in order to study the effects of melanin on norharman-induced toxicity. In this model, melanin was found to attenuate the toxicity induced by

low concentration of norharman. Following a high concentration of norharman, melanin still attenuated necrosis but also gave rise to a high level of cellular stress and apoptosis. We are currently examining the uptake and effects of an algal neurotoxin BMAA (beta-N-methylamino-L-alanine) in the brain of rodents. BMAA is a non-protein amino acid that is produced by cyanobacteria. This neurotoxin has been suggested to contribute to neurodegenerative disease.

Olfactory transfer of drugs (Elena Piras)

The olfactory pathway is a potential route of delivery of therapeutic agents that do not easily pass the blood-brain barrier. The olfactory neurons have direct contact with the external environment via dendrites in the nasal mucus and with the brain via axons that reach the olfactory bulb without synaptic connections. The olfactory transfer of CNS-active drugs into the brain is a novel principle for drug delivery. We have demonstrated a transfer of morphine and dopamine via the olfactory pathways to the brain following intranasal administration in rodents.

Members of the group during 2007

Eva Brittebo, Professor
Nils Gunnar Lindquist, Adjunct Professor
Helén Andersson, PhD Student
Oskar Karlsson, PhD Student
Elena Piras, PhD Student

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Genetic Toxicology

Björn Hellman

When testing the potential DNA-damaging effects by medical drugs and other chemicals, the test systems are generally based on experimental animals, bacteria or various kinds of transformed cells. For the safety evaluation it would be of advantage if healthy cells from humans could be used instead, since they have a normal and stable set of chromosomes. In case of using primary cultures of human lymphocytes, it is hardly possible to use a blood sample on more than one single testing occasion. This will often give varying results from different testing occasions, since new blood samples (in general from different donors) have to be taken all the time. Our extended-term cultures of lymphocytes allow one single blood sample to be used for up to 50 different experiments. We measure the DNA-damage with the so called Comet Assay, which is a relatively quick, simple and cheap method for evaluating DNA-strand breaks in individual cells. The main target of our project is to improve the risk assessment regarding exposures to genotoxic agents.

From point of view of risk assessment, it is important to differ between genotoxic carcinogens and other substances that increase the risk of cancer by other mechanisms. In the case of drug-induced oxidative DNA-changes, for instance, one can distinguish two different main groups of substances: those who cause various types of reactive oxygen radicals in the cells directly and those who cause oxidative stress indirectly, as a consequence of general cytotoxicity. The research of recent years has also shown that the DNA repair has a great impact on whether the DNA-damage is manifested as a mutation or not, and there is reason to believe that there is a great variation in individual sensitivity to genotoxic agents, due to individual differences in DNA repair, metabolic bioactivation/detoxification pattern and/or other defense mechanisms in the cells. All those aspects are studied in this project.

In a recent project, supported by SIDA SAREC, we are also evaluating the genotoxic and antigentoxic effects of some plant extracts used in traditional medicine in Ethiopia. In this project we are also using an in vitro version of the micronucleus test to evaluate potential clastogenic effects.

Members of the group during 2007

Björn Hellman, Associate Professor; Ph.D.; Senior Lecturer and Study Director in Toxicology
Jemmal Demma; M.Sci. in Pharmacy; Ph.D. student
Lena Norgren; Laboratory assistant

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Agencies that support the work/Funding 2007

SIDA/SAREC: 151 000 SEK

Undergraduate Teaching

In July 2007, a new law for higher education was put into practice with a view to better harmonize Swedish higher education with educational systems within Europe. The change is in line with those ongoing European collaborative development processes that are described in the Bologna declaration. The so-called Bologna process aims to increase student and teacher mobility, enhance employability and overall to strengthen the competitiveness of European education. In the new Swedish educational system, undergraduate studies are divided into two levels, that is, a student starts at the basic level (3 years of studies) and may thereafter continue at the advanced level (1-2 years). After the advanced level, the student may continue graduate studies at the research level. During the past year, the department staff made a considerable work effort to adjust programmes and courses to the new system according to the new Higher Education Act. In the new system, 1 hp (higher education credits "högskolepoäng") equals 1 credit in the ECTS (European Credit Transfer System). The grading system within the Faculty of Pharmacy was also revised in 2007. Examinations are from July and onwards graded with three grades, U (not passed), G (passed) and VG (passed with distinction).

The Department of Pharmaceutical Biosciences is responsible for teaching at two main programmes, the Bachelor of Science in Pharmacy programme (180 hp) and the Master of Science in Pharmacy programme (300 hp). Completed studies on the Pharmacy programmes provide the necessary theoretical and practical competence that is required to apply for a licence as a Pharmacist, either Receptarie (after the Bachelor programme) or Apotekare (after the Master programme). In addition, teachers within the department are involved in teaching at the Master of Science in Chemical Engineering, with specialization in drugs, and the Biomedical programme. The main subject taught at the department is Pharmaceutical Biosciences that comprises a large number of courses: Drug development and Drug usage, Drug metabolism and safety, Gene technology, Immunology, Infection Biology, Microbiology, Molecular Biology, Pharmaceutical Biochemistry, Pharmacology, Pharmacokinetics, Pharmacotherapy, Physiology, Toxicology. A mix of traditional teaching and problem-based learning with lectures, laboratory sessions, seminars, group work and computer sessions characterizes the teaching. In addition, the teachers are involved in interdisciplinary training in laboratory practice, communication skills and professional development.

Besides the mandatory courses at the programmes, the department gives many elective courses on the advanced level. The courses attract a large number of students, not only Pharmacy students, but also other students showing the proper prerequisites in biosciences. These courses mirror research profiles within the department, such as bioinformatics, drug metabolism and safety, drug dependence, and pharmacokinetics. The teachers also instruct in undergraduate projects. These projects comprise 15 hp or 30 hp and are examined by an oral presentation as well as a written report. Most of the projects within biosciences involve laboratory-based projects in which the student is involved in ongoing research projects.

Uppsala March 5, 2008

Ingrid Nylander

Awards and Appointments 2007

1. The Bioclipse system developed by professor Jarl Wikberg's team in collaboration with a group of international developers was awarded the Special Prize of the jury at the international contest for free software, Trophees du Libre, 2007 which was held in November 2007 in Soissons, France.
2. Dr Mats Magnusson received the Rosenö award for best Swedish thesis in the area of pharmacokinetics, pharmacodynamics, drug metabolism and clinical pharmacology.
3. PhD-student Radojka Savic was selected as Lewis Sheiner Student Speaker at the 16th PAGE conference in Copenhagen, Denmark.
4. Dr Mathias Hallberg was awarded the Benzelius award at the Royal Swedish Society of Science for the thesis "Anabolic androgenic steroids; Effects on neuropeptide systems in the rat brain"
5. Professor Fred Nyberg was awarded "Karl Johan Öbrink lecturer" 2007 at BMC, Uppsala University.
6. Dr Camilla Svensson was awarded the Pedagogical prize 2007 from The Pharmaceutical Student Union.

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