UNIVERSITÄT SALZBURG

Research Topics at the Department of Molecular Biology

Laboratory impressions...

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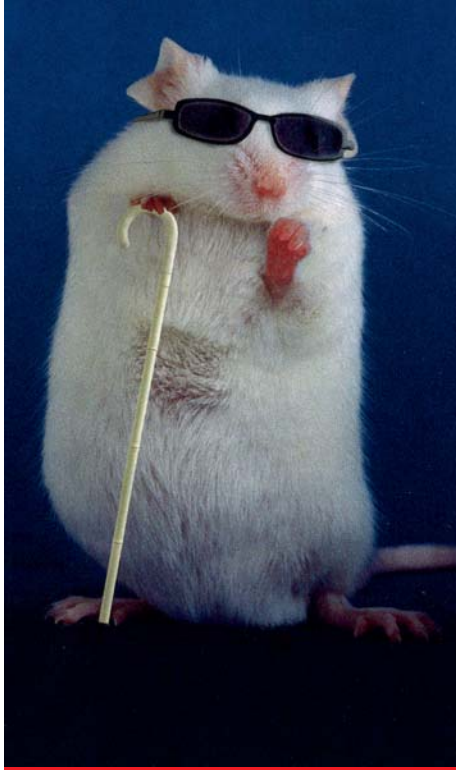
Welcome to the Department of Molecular Biology

Research-driven teaching at an international level is one of the hallmarks of academic training. The Department of Molecular Biology warrants these ambitious standards by introducing the students to a variety of current research projects, enabling them to actively participate in and propel these projects using state-of-the-art methods.

Sixteen research groups at the department share their expertise in the areas of allergology, immunology, structural and chemical biology, bioanalytics, proteomics, (epi-)genomics, tumour biology, stem cell biology, microbiology, glycobiology, molecular plant biophysics and biochemistry. Within **interdisciplinary teams** we tackle current challenges in allergy and immunology, cancer, and bio-nano-interactions from multiple angles. We **publish** our research results **in internationally peer-reviewed journals** and further translate them into practice by diverse **cooperations** together **with clinical and industrial partners**.

Salzburg, August 2016

Hans Brandstetter, Head of the Department of Molecular Biology



Research and Teaching at the Department

The Department strives to offer **teaching and research activities at the forefront of science**. To accomplish this ambitious goal, we organize **research and teaching as closely interconnected processes** that enable their constant refinement and improvement. Students and staff work hand in hand in their implementation.

Within the biological undergraduate curricula our department currently runs two bachelor (**biology and molecular biosciences**) and two master (**molecular biology and medical biology**) programmes; we further contribute to the bachelor in **engineering** program, to the master programmes **ecology & evolution, chemistry & physics of materials**, and **data science**, as well as to **secondary teaching degree programmes in biology and chemistry**.

Finally, we offer most **competitive graduate training programmes**. These programmes reflect the international reputation of the individual laboratories of our department as well as the quality of the PhD students that qualify for them in international selection processes. Our department succeeded in several structured graduate programmes, including an FWF-funded programme on **Immunity in Cancer and Allergy (ICA)**; a Christian Doppler Society-funded programme on **Biosimilar Research**; two Doctoral School PLUS programmes on S**tructure, Function and Regulation of Biomolecules** and **Statistics and Applied Data Science**; and several European and nationally funded training networks and individual PhD programmes, all of which witness the **cutting edge research** that is carried out at our Department. Highlights of the research programmes are given on the next pages.

The department's success of the last decade would not have been possible if not for the success to acquire significant funding that is necessary to establish, run and maintain **excellent research infrastructure**. We are particularly thankful to the State of Salzburg for endowing two chairs in structural and chemical biology and supporting the infrastructure in the Christian Doppler Laboratories; to the University of Salzburg for enduring support within the priority program; and the ongoing support by several additional public and private partners. Their trust in us is our responsibility and commitment.

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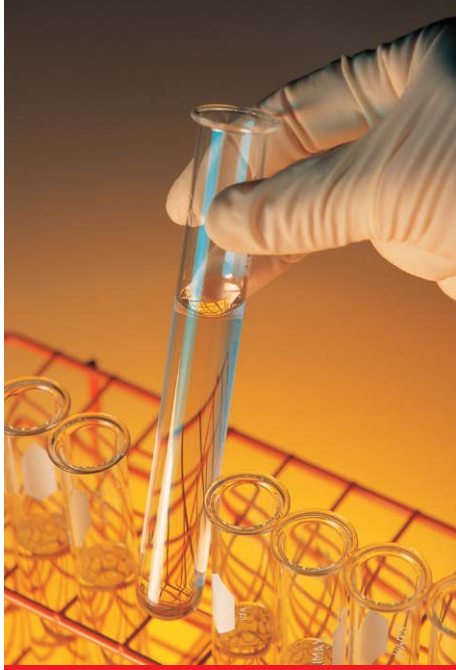
Overview of Research Topics of the Department

The sixteen research groups of the department cover a **broad variety of expertise**, ranging from immunology, allergology, structural biology, tumor biology, genomics, glycobiology to bioinformatics, molecular plant biophysics and biochemistry, bioanalytics, biological chemistry and microbiology. In a **highly mutidisciplinary approach** and by employing cutting-edge technologies the department addresses urgent and current topics in the fields of **allergy and immunology, tumor biology, and nanotoxicology**.

The **division of allergy and immunology** works on the development of new tools for allergy diagnosis and allergen-specific immunotherapy, prophylactic vaccination against allergies, signal transduction within cells of the immune system, the effects of environmental pollutants on immunity, and molecular mechanisms associated with allergy and asthma. In the **division of tumor biology**, researchers try to understand the mechanisms set in motion by oncogenic signaling pathways during the initiation and growth of human malignancies. On the therapeutic side, cellular mechanisms of photodynamic tumor therapy are investigated.

The **divisions of structural biology and bioinformatics** and **chemistry and bioanalytics** are specialized on the study of molecular details and mechanistic principles of proteins central to allergy or cancer. Genomic, proteomic, and metabolomic approaches are implemented to reveal a global view on the molecular networks involved in biological processes. Interactions of host cells with microbial pathogens are studied in the **division of microbiology**. Finally, biological functions of glycans in the regulation of cell differentiation, signal transduction, control of apoptosis and in the development of malignant cancers as well as the polarity in biological systems such as pollen tubes are studied in the **departments of glycobiology** and **molecular plant biophysics and biochemistry**.

Candidates interested to join the research activities will obtain a **multidisciplinary training in state of the art techniques**. The department provides access to the necessary technological infrastructure and expertise that is necessary to conduct biological research on an internationally competitive level.



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Research and Teaching Infrastructure of the Department

Important technologies and large-scale instruments utilized at the Department include, *e. g.*: (1), cell counting; (2), fluorescence microscopy; (3), fluorescence spectroscopy; (4), patch clamp electrophysiology; (5), chip-based trascriptomics; (6), X-ray crystallography; (7), nuclear magnetic resonance spectroscopy; (8), high-performance liquid chromatography and mass spectrometry; (9), fluorescence-activated cell sorting; (10), quantitative methylation analysis by pyrosequencing.

Focus Area “Allergy-Cancer-BioNano Research Centre – ACBN”

ACBN was established by the Paris-Lodron-University of Salzburg in 2015 out of the original priority program “BioScience and Health” which had been founded in 2003. The declared aims of the program are performance of **excellent basic science and translational research** with high international visibility and recognition. Ten working groups of the Department as well as two more of the Chemistry and Physics of Materials Department investigate the molecular and cellular basis of different diseases and disease mechanisms.

Current research collaborations focus on the topics **allergy, immunology, cancer research, bio-nano interaction, and structural biology**. The basic research performed in ACBN should help to **better understand the molecular and cellular basis of diseases such as cancer or allergy**, in order to develop better **tools for their diagnosis and therapeutic treatment**. Synergistic use of knowledge, expertise, competence, and infrastructure facilitates multidisciplinary approaches to scientific problems and ideas, and leads to a constructive and inspiring working environment. This also impacts our young scientists, who thus derive great benefit and learn to embed their projects into a broader context. Finally, also **teaching** very significantly **benefits from the expertise, research tools, and infrastructure available in the priority program**, thus providing a solid and real-life oriented basis for the state-of-the art education of students in the different study programs of biology at the University.

Another important aspect of the continuous development of the focus area is the preparation and **founding of larger research structures**. ACBN members were successful in recruiting third party funding for individual projects but also in the establishment of national and international large-scale projects, such as Christian Doppler Laboratories, joint projects within European Framework Programs, and national and international training networks/doctoral colleges. Recently, members of ACBN founded the “Cancer Cluster Salzburg – CCS”, a collaboration network of basic scientists and clinicians with a unique focus on tumor microenvironment and tumor stem cells.

Up to the present time, ACBN expanded step-by-step and developed a clear and distinct research profile, thus being a good example of the successful implementation of focus areas by the University of Salzburg.

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Doctoral College Immunity in Cancer and Allergy – ICA

The **mammalian immune system** acts as a complex surveillance system ensuring to **distinguish between self and non-self**, and between **harmless and dangerous** events approaching from outside or inside of the organism. Moreover, it interacts with growth, differentiation and death of cells and tissues, and thus maintains the homeostasis and the integrity of our body.

The doctoral program **"Immunity in Cancer and Allergy - ICA"** focuses on two pathologies of the immune system, i.e. the **overwhelming allergic immune response** and the **inefficient immune response against certain tumors**. Both diseases are a growing concern and there is an urgent medical need to elucidate the underlying mechanisms for the development of new therapies. The scientific goal of the program is unraveling the cellular and molecular immunological mechanisms and pathways and to develop rational and molecule-based strategies for the treatment of these diseases. For this purpose, ICA selects **excellent graduate students from all over the world**, provides an intellectually stimulating environment, excellent instrumental and methodological infrastructure and ambitious scientific projects.

Nine work groups of the Department of Molecular Biology led by Fritz Aberger, Hans Brandstetter, Albert Duschl, Fatima Ferreira, Iris Gratz, Christian Huber, Angela Risch, Josef Thalhamer and Silja Weßler, as well as Richard Greil and Tanja Hartmann from the Salzburg Cancer Research Institute guarantee **high quality research and training**. Their overall aim is to **create an interactive environment, including a broad range of scientific, technological and methodological expertise**. Common and overlapping scientific interests of the college members build a solid basis for solving complex questions in the field of allergy and tumor research that are beyond the capability of the individual groups.

ICA has meanwhile been successfully evaluated twice by international Evaluation Commissions of the Austrian Science Fund, and recently the program has been granted for a further period of three years. At present ICA comprises eleven faculty members and twenty-four fully financed doctoral students from seven different countries.

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Cancer Cluster Salzburg-Center for Tumor-Microenvironment Communication

CANCER CLUSTER SALZBURG
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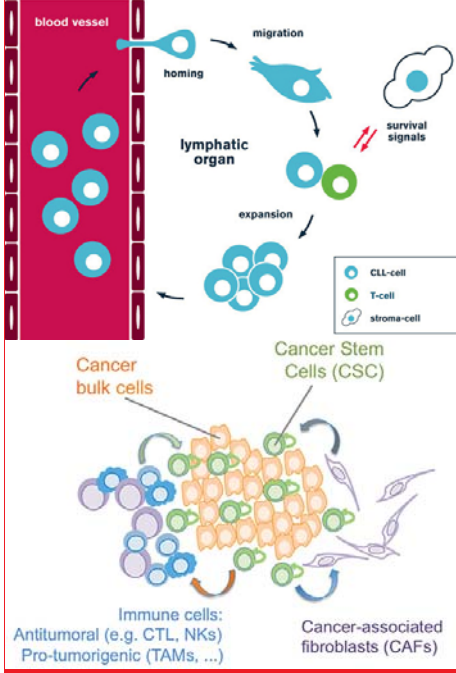
By 2030 the global cancer burden is projected to reach more than 21 million new cases and 13 million deaths per year. Despite considerable progress in the fight against cancer, **malignant diseases are becoming a global pandemic** and the development of **innovative and more efficient therapies** is a major challenge in biomedical research of the 21st century. **Understanding the highly complex nature of cancer** on a molecular and cellular level is key to targeted and improved therapies. This requires joint, multi-disciplinary efforts of molecular cancer biologists and clinicians to identify the causes of malignant cell growth and to swiftly translate basic knowledge into clinical application.

CCS has been founded as a contractually agreed consortium of 16 interdisciplinary and internationally recognized research groups of the Department of Molecular Biology of the Paris-Lodron University of Salzburg (PLUS), the Salzburg Cancer Research Institute (SCRI), and the Paracelsus University Clinics Salzburg (SALK/PMU). CCS represents an **innovative and dynamic research and training network** of molecular cancer biologists, oncologists, oncosurgeons, pathologists, immunologists, and systems- and structural biologists, including Fritz Aberger, Hans Brandstetter, Chiara Cabrele, Iris Gratz, Jutta Horejs-Höck, Christian Huber, Angela Risch, and Silja Weßler from the Department of Molecular Biology. Eight CCS groups are also members of the FWF-funded International PhD program Immunity in Cancer and Allergy, thereby providing to young and highly talented scientists an excellent training environment in the field of molecular and immunological cancer research.

The scientific focus of CCS is on the **understanding of intricate cellular communication processes between malignant cells, their microenvironment and the immune system**. Unraveling the molecular details of tumor-microenvironment interactions, in particular the mapping of oncogenic driver signals within the complex tumor micro-milieu, including the immune system, will provide new opportunities for improved therapeutic concepts tackling the most imminent challenges in oncology such as relapse, metastasis and drug resistance for the patients benefit.



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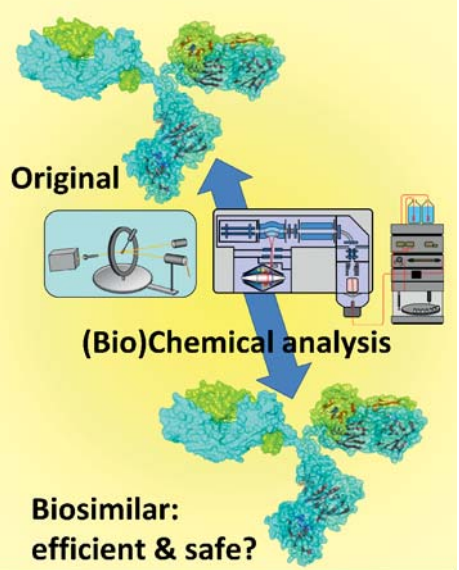

Christian Doppler Laboratory for Biosimilar Characterization

Biopharmaceuticals, or biologics, are medicines produced from living organisms using biotechnological techniques. They represent already one third of the new drugs currently under development. **Biosimilars are new versions** of existing biopharmaceuticals (originators) following loss of patent protection. In contrast to classical chemical drugs, such as Aspirin® or Penicillin, therapeutic proteins represent **very complex, macromolecular compounds** which must be thoroughly qualified with regard to their quality, efficacy, and safety before their application in therapeutic medication. The more **physical, chemical, and biological data** we can obtain for the produced drugs, the higher is the certainty that only desired effects and no adverse effects occur during the therapy.

Upon joining the expertise in **protein production, characterization and analysis, structural biology, and synthetic chemistry** existing in the groups of Hans Brandstetter, Chiara Cabrele, Gabi Gadermaier, Christian Huber, and Hanno Stutz at the Department, the Christian Doppler Laboratory of Biosimilar Characterization aims at developing innovative characterization tools for the efficacy and safety of therapeutic proteins and their transfer to the industry. **Sandoz,** one of the biggest pharma companies in Austria with prominent sites in Tyrol, Austria, as well as **Thermo Fisher Scientific**, based in Massachusetts, USA, one of the largest global suppliers of laboratory equipment and scientific instruments, joined as industry partners to put the Christian Doppler Laboratory on a solid and application-oriented basis.

Complex biomolecules such as **therapeutic proteins** feature many **intrinsic properties that are essential for the safety and efficacy** of the medical agents. The **primary structure** as well as **posttranslational modifications** (including glycosylation, oxidation, deamidation) are usually determined by high-performance liquid chromatography hyphenated to high-resolution mass spectrometry. Chemical biology approaches are implemented in order to provide standardized material mimicking relevant protein modifications Other aspects of protein structure, such as **protein folding or biological function** are addressed by means of X-ray crystallography, capillary zone electrophoresis, circular dichroism spectroscopy, infrared spectroscopy, or biological assays based on the treatment with specific enzymes (or enzyme cascades) or on the binding of RNA-based aptamers.

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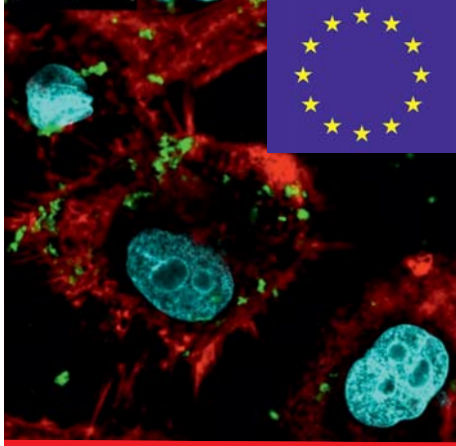
Projects Supported by the European Union

Research projects funded by the EU create **thematic networks between European Researchers**, address **topics of transnational interest** and support the **career development** of young scientists. The European research agenda is defined by framework programs, which usually run for 7 years. The presently active program, called Horizon 2020, is the 8th framework program. Researchers from the department have been strongly involved in previous programs and are also involved in the ongoing Horizon 2020 initiatives.

Albert Duschl has recently coordinated the projects **NanoTOES** and **NanoEIS**, led a work package in **NanoValid**, and is at present active in **HUMUNITY** and **PANDORA**. These projects focus mainly on **aspects of bio/nano interaction, in particular following inhalation of nanoparticles**. The human immune system is particularly sensitive to disturbances by nanoparticles that are recognized and frequently taken up by immune cells. This may lead to adverse reactions, or, if the particles are harmless, to tolerance. The team has established **robust tests for nanosafety**, which can be used to identify problematic particles early on. Specific effects on the immune system may open new perspectives to use such materials in nanomedicine, which is explored for example with respect to allergic diseases. Christian Huber has been involved in the project **PREDICT IV** (Profiling the Toxicity of New Drugs) and **MARINA** (Managing the Risks of Nanomaterials), which were both focusing on the develoment of non-animal-based **testing systems for the toxicity of drugs and nanomaterials**, respectively, involving holistic **proteomics, metabolomics, and transcriptomics** approaches. Fatima Ferreira-Briza participates in the project **BM4SIT** - An Innovative Causal Therapy for Allergy. The **hypoallergenic birch pollen vaccine BM4**, which was developed in this lab, is currently being clinically evaluated in BM4SIT, a project financed by the European Union. Therefore, a therapy concept is being developed, which will allow a reduction of the number of injections necessary for the treatment of allergic disease.

Overall, the support from the European Union is an important asset for the Department. It increases our international visibility, allows to address topics that are accessible only by working within large networks, and it gives our young colleagues excellent career perspectives by fully involving them in international research efforts at an early stage.



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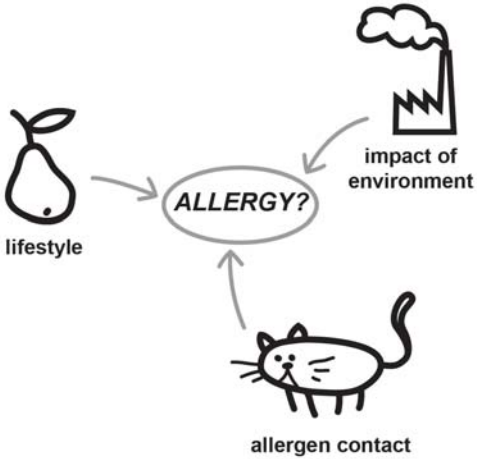
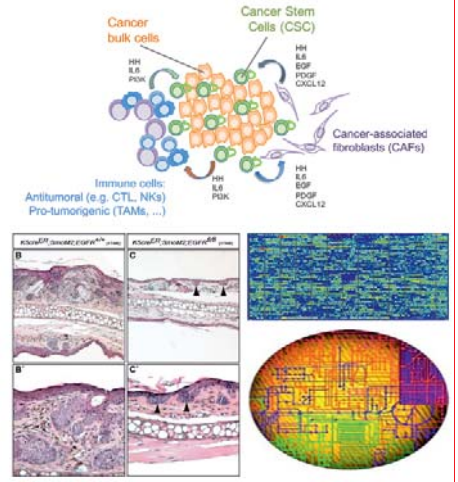
Sparkling Science and Citizen Science Projects

In Sparkling Science projects funded by the Federal Ministry of Science, Research and Economy, **scientists work side by side with pupils to address state-of-the-art research questions and thus encourage young scientists**. The Sparkling Science project **BIO-KoSMoS** understands itself as link between biotechnology, medicine and modern art. Allergens are being produced using novel technological platforms. The development of such platforms is accomplished in collaboration with high schools using fluorescent proteins as test material. The fluorescent proteins themselves are then further processed as pigments to manufacture BioArt inks and dyes. Projects like BIO-KoSMoS ensure inspiring and **sustainable science education in a close cooperation between the university and high schools** (www.biokosmos.org). The allergy project **ALRAUNE** investigates allergen **exposure and IgE sensitization of 500 pupils from Salzburg**. Our study shows that 53% of pupils carry allergenic antibodies and the most common allergy elicitors were grass and tree pollen as well as house dust mites and cat hair. Lifestyle factors like genetic predisposition, smoking, high hygiene and lack of farm exposure significantly influence the development of allergies.

Pupils are directly involved in the project and execute allergen sampling and laboratory experiments, develop hands-on material for schools and public relations, practice peer-to-peer teaching and present the scientific results life on stage in a Science Slam.

Citizen Science (Open Innovation) describes the **opening of science to the general public**. Interested **citizens provide valuable input** and know-how for research projects. Within the Citizen Science Project **ALRAUNE – Tracking allergies**, pupils and interested citizens develop an allergy questionnaire aiming to identify facts why allergies are still on the rise.





Team Fritz Aberger
Division of Molecular Tumor Biology

Cancer is a leading cause of death worldwide. Despite considerable progress in the treatment of cancer patients, the incidence and death toll are globally increasing, urgently calling for **intensified research to better understand the molecular basis** of this disease and to identify improved strategies and drugs for personalized therapy.

As partner within national and international research consortia and active founding member of the Cancer Cluster Salzburg (CCS), the Aberger lab applies a **combination of in vivo cancer models and state-of-the art cellular, molecular and systems biology approaches**, to **decipher druggable molecular pathways** such as **Hedgehog/GLI signaling** and to unravel novel molecular targets for cancer therapy. A central research focus of the group is on the analysis of **pathways controlling the malignant potential of cancer stem cells (CSC)**, a small subpopulation of tumor cells responsible for cancer growth, metastasis, drug resistance and disease relapse. In particular, the lab tries to understand the interaction of CSC with the **tumor microenvironment** including the **immune system**.

The core competence of the group is the **functional analysis and cross-talk of oncogenic Hedgehog signaling and GLI transcription factors with other cancer pathways** in malignant cells and the tumor environment. By bringing together the expertise of molecular tumor biologists and clinicians within the Cancer Cluster Salzburg, the Aberger group aims to translate preclinical studies to clinical application for better therapeutic outcome.

As major contributions, the Aberger lab has recently deciphered oncogenic signals and novel drug targets responsible for malignant disease progression. In c**ollaboration with partners from the biotech industry**, the group works on the development of novel **drugs for improved anti-cancer (immuno)therapies,** including innovative inhibitors of malignant cancer stem cells.

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Team Hans Brandstetter
Division of Structural Biology & Bioinformatics

The research of our team focuses on **proteolytic enzymes**, their cofactors, substrates and inhibitors. Proteolytic enzymes are master switchers in health and disease. By their highly specific recognition and modification of protein substrates, **proteases serve as signaling and decision matrices**. Of particular interest to us are proteolytic actions in the field of immunology/allergy, blood coagulation, and cancer.

We employ a broad spectrum of **biochemical, biophysical and computational techniques** to **characterize the molecular function of important target proteins**. Most importantly, we use **x-ray crystallography** to determine the three-dimensional architecture of the molecular targets. The structural information guides us to rationalize and experimentally test hypotheses about possible molecular mechanisms of action as well as their significance in the molecular, cellular and systemic context.

Questions of immunological interest are linked to the **cellular pass control** which is implemented *via* antigen processing and presentation. Each cell has to identify itself towards the immune system by presenting intracellular peptides at the cell surface. Despite its enormous efficiency, some **harmful cells escape the screening** (pass control) of the immune system, e.g., by presenting unsuspicious peptides only. On the other side, **allergens,** which are *per se* harmless proteins, **can trigger excessive immune reactions**. We investigate the complex protease machines involved in antigen processing. A detailed understanding promises treatment options against immunological and infectious diseases, and also tumors.

The (innate) immune system is multiply linked to the **blood coagulation system** which primarily serves to stop life-threatening bleeding without causing fatal vessel occlusions (thrombosis). For this purpose nature employs a **molecular proofreading principle** that requires simultaneous and concerted molecular actions. We investigate disharmonies in these molecular orchestra that lead to bleeding disorders such as hemophilia.

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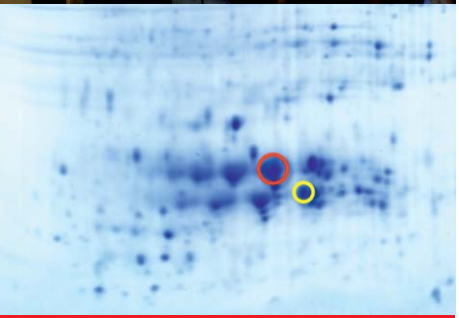

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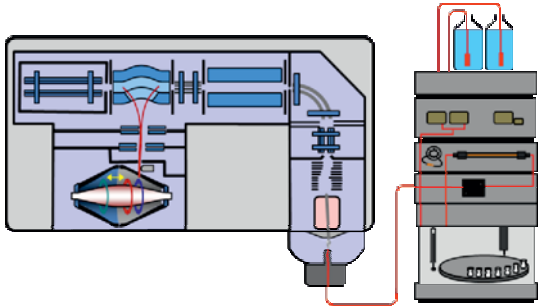
Team Peter Briza
Division of Allergy and Immunology

With the development of modern tandem mass spectrometers, **proteomic research** became an increasingly important tool to identify and characterize proteins in complex samples. Several complementary approaches are possible. Proteins are extracted from cells or organelles, digested with protease and the resulting peptides are separated by reverse phase HPLC and analyzed by a directly coupled tandem mass spectrometer. Another option is to separate the extracted proteins by **one- or two-dimensional gel electrophoresis**, digest protein spots with protease and analyze the peptides by MS. Tandem MS instruments are able to simultaneously acquire peptide masses and peptide fragmentation patterns. With this information, **proteins can be unequivocally identified in protein data bases**. Sequence information obtained for unknown proteins can be used for cloning the corresponding genes. Our group uses a state-of-the-art Quadrupole-Orbitrap mass spectrometer with electrospray ionizatiion, optionally coupled with a capillary HPLC.

Using the methodology described above, we try to **characterize and verify purified allergens**, as well as **identify unknown proteins or protein isoforms that induce type I allergies in allergic patients**. Our main focus are allergens from tree (birch and selected trees), weed (mugwort, ragweed) pollen and food (peanut). Our work is done in close cooperation with the group of Fátima Ferreira.



Team Chiara Cabrele
Division of Chemistry and Bioanalytics

Our research focuses on the investigation of the chemical, biophysical and structural properties of proteins, aiming at the deep understanding of their mode of action and with the long-term goal to exploiting the acquired knowledge to design and develop artificial molecules as positive or negative modulators of protein activity. In particular, we are interested in small regulatory proteins that play a role in cancer and vascular diseases.

Proteins are key biomolecules in many cellular processes including the activation of DNA transcription, the transport of messenger RNA from the nucleus to the cytoplasm, the biocatalysis of reactions (which otherwise would be slow and unspecific), the regulation of signaling cascades. Despite their diversity, all these processes share a fundamental mechanism, namely **protein molecular recognition**. Proteins may recognize themselves or other proteins as well as different classes of molecules (e.g. DNA, RNA, glycans) by means of specific non-covalent interactions such as hydrogen bonds, electrostatic and hydrophobic contacts. It is thus important to characterize the non-covalent network that allows biomolecules exerting their function. This implies a deep sight into the structure and dynamics of proteins by using spectroscopic techniques (circular dichroism, fluorescence, nuclear magnetic resonance), which in some cases requires the use of **chemically modified proteins.**

Organic chemistry methods applied to proteins allow reproducing, mimicking and manipulating these very interesting biomolecules. For example, the preparation of **proteins containing post-translational modifications** (e.g. phosphorylation, acetylation, methylation, glycosylation) provides the tools to evaluate the structural and functional importance of these modifications that are essential to regulate the activity of proteins in the cells. Furthermore, the mimicry of protein interfaces by means of synthetic scaffolds leads to **artificial, proteolitically stable modulators of protein-protein interactions**, which are of significance for the development of drug-like molecules.

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Team Albert Duschl / Jutta Horejs-Höck
Division of Allergy and Immunology

At the center of our interest are **effects of various factors on the human immune system**. We are constantly subjected to a multitude of natural environmental stimuli (bacteria, viruses, fungal spores, allergens, etc.), but anthropogenic factors are present as well and may be increasing (fine dust, nanoparticles, exhaust gas, etc.). The current boom in nanotechnology may create safety hazards, but also promises groundbreaking applications, including medical ones. Against this background, **investigations into nano-bio-interactions** and into **molecular mechanisms of regulations** for different immune cells have developed into our major areas of research. Some *hot topics*:

Nanosafety – nanotechnology applies extremely small materials (1-100 mm), which have novel and attractive properties based on low mass and high surface area, allowing new technical applications. Due to their small size and their high surface reactivity they are able to penetrate body barriers, like airways, lung and gastrointestinal tract, which may induce both toxic and immuno-modulating responses. Biological effects can carry risks, but may also be useful for medical applications.

Nano-Bio-Interactions – the highly reactive surface of nanomaterials causes quick and often rather stable binding of biological molecules, mainly proteins, which affects reactions of the body. This property may be used for intentional transport of proteins and other substances; however, binding to nanosurfaces can alter structure and function of proteins. Consequences for immunity are under study.

Interaction between unspecific and specific immune response – Dendritic cells recognize foreign substances via *„pattern recognition“* receptors, take up antigens and activate T-cells, which induce now an immune response that will either result in defensive actions, or in the establishment of tolerance. We focus on molecular mechanisms involved in activation of dendritic cells, since they play a key role in deciding how the immune system will react to non-self substances.

Team Fátima Ferreira-Briza / Michael Wallner
Division of Allergy and Immunology

Type I allergic reactions comprise a wide range of IgE-mediated diseases, which affect more than 20% of the population and represent a major health problem worldwide. The primary interest of our group is the development of new tools for **allergy diagnosis** and for safer and more efficient **vaccines for allergy**. Our research focuses on **allergic reactions triggered by pollen** (birch, mugwort, ragweed), **mites,** and **foodstuffs** (apple, peach, celery, nuts). One major goal of our research is to understand why some proteins are allergenic and others not.

The identification of novel allergen molecules is essential for improving molecule-based allergy diagnosis. For this purpose, we have established methods for cloning and **recombinant production** of allergens in heterologous expression systems like bacteria, yeast and plants. The identity and secondary structure elements of the purified allergens are routinely characterized using **physicochemical** (gel electrophoresis, mass spectrometry, amino acid analysis, circular dichroism, dynamic light scattering) and a wide array of **immunological methods** (ELISA, mediator release assays, simulated antigen processing assays, uptake by antigen presenting cells, immunization models).

Well-characterized allergen molecules are used for diagnostic studies including the development of **allergen microarrays**. Another goal of our group is the generation of safe and efficient tools for allergy vaccination. Using knowledge-based approaches, we have developed **hypoallergenic molecules** of important pollen and food sources (e.g. birch pollen, peach). Before clinical trials, the **efficacy of candidate vaccines** is usually tested in murine models of allergic sensitization. The **hypoallergenic birch pollen vaccine** BM4, which was developed in our lab, is currently being clinically evaluated in the framework of a project financed by the European Union. As member of the Christian Doppler Laboratory for Biosimilar Characterization, we want to generate novel structural and immunological tools for comparison of originator biopharmaceuticals and biosimilars.

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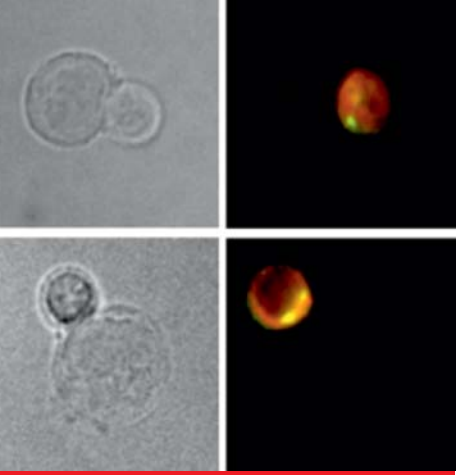

Team Iris Gratz / Gertrude Achatz
Division of Allergy and Immunology

Allergy and autoimmunity are chronic and debilitating conditions for which current treatment approaches are unsatisfactory. These inflammatory diseases are caused by immune cells attacking **harmless (self-)antigens in host tissues**, which they are supposed to protect. However, further basic research is required to fully understand the disease mechanisms and immune-regulatory processes in the target tissues.

Our research aims to **elucidate the mechanisms that control the balance of immune activation versus tolerance** because this balance ultimately determines the outcome and severity of disease. Understanding these mechanisms will help to develop novel therapeutic strategies. In our studies of autoimmunity and allergy we focus on immune-regulatory processes of two major epithelial body surfaces, skin and lung.

Our main research questions are: 1. The **role of Foxp3+ regulatory T cells** (Treg) in inflammation, their generation and recruitment to the target tissues. 2. **Immune cell stability and memory formation of T and B cells** with an emphasis on apoptotic processes and their regulation by cytokines. 3. **Modulation of human immune responses** in clinical settings such as gene therapy

In our projects we study cellular processes of T and B cell differentiation, memory formation, and IgE-regulation. For these studies we use **innovative mouse models and humanized mice**, which we manipulate with genetic and biomolecular tools. We combine the use of these **transgenic and knock-out mice** with the application of biologics, antibodies and small molecules to manipulate immune cells in vivo.

In all of our approaches we focus not only on the phenotypes of selected lineages but also on the **crosstalk between immune cells of the innate and adaptive immune system**. We are particularly interested in the **interaction between T cells and antigen-presenting cells (APCs) or B cells**.

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Team Christian Huber / Hanno Stutz
Division of Chemistry and Bioanalytics

The research focus of our team regards the development and application of **analytical workflows** to address biological questions in the fields of **protein, proteome, metabolite, and metabolome** (and eventually transcriptome) **analysis.** Samples comprising **cultured cells, tissues, or biological fluids** are processed and their constituents of interest (proteins or metabolites) isolated for further determination. The analytical methods are primarily based on instrumental, bioanalytical separation methods (**liquid and gas chromatography, capillary electrophoresis**) in combination with **mass spectrometry** (time-of-flight-, triple-quadrupole-, linear ion trap-, and Orbitrap mass analysis). Because of the enormous amount of generated raw data, we collaborate with bioinformaticians and statisticians in order to properly interpret the experimental data and put them into a biological context.

The major goal of our work is the collection of information about **changes in protein or metabolite concentration** that are **caused by stimulation** of cell models (cancer stem cells, dendritic cells, monocytes, hepatocytes, lung epithelial cells) upon treatment with drugs, nanomaterials, or **by diseases such as allergy or cancer**. These changes allow us drawing conclusions on the **biochemical pathways and mechanisms** involved in disease or toxic effects of drugs and nanoparticles. In such experimental setups, we use, e. g., dendritic cells isolated from human blood to study the effects of allergens on the immune system.

In a second focus area we collaborate with the **pharmaceutical industry** (Sandoz) and the **laboratory supplier industry** (Thermo Fisher Scientific) in the **Christian Doppler Laboratory for Biosimilar Characterization**. Here, we use our expertise for the in-depth protein characterization (peptide mapping, sequencing, determination of impurities, glycosylation, oxidation, and deamidation) to aid the industry in establishing **workflows that guarantee the safety and efficacy of their biopharmaceutical drug products**. This research focus requires intensive collaboration with groups of the department having expertise in protein production, chemical protein modification, structural biology, and biochemical protein characterization.

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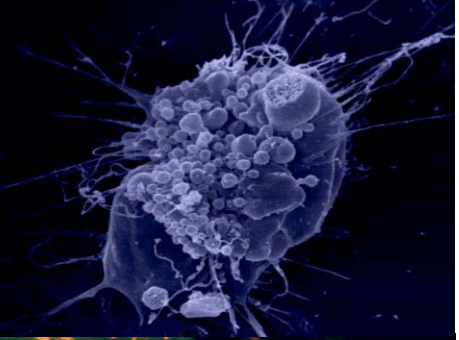

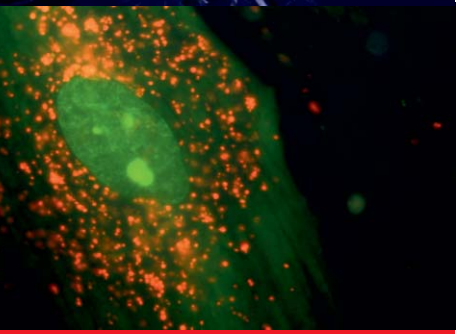

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Team Barbara Krammer
Division of Molecular Tumor Biology

In research we focus preferentially on **molecular and cellular mechanisms of the Photodynamic Tumor Therapy (PDT) and Diagnosis**. PDT combines a selective uptake of a photosensitizer to tumor tissue with irradiation using visible light. By this, the photosensitizer is activated triggering either fluorescence or physico-chemical reactions with other molecules, mainly molecular oxygen. The **fluorescence marks the tumor (clinical diagnosis)** and the chemical reactions lead to **formation of reactive oxygen species** at the target sites **resulting in cell death.** Thus, malignant tissue at inner and outer surfaces of the body can be removed without inducing major side effects or mutagenicity.

Since we are interested in investigating the mechanisms accounting for the observed clinical effects and in developing improved protocols, we use a large variety of in-vitro methods ranging from **spectroscopic measurements over cell based assays to molecular biology techniques** utilizing *inter alia* a flow cytometer, microplate reader, fluorescence microscope, fluorescence spectrophotometer and a real-time PCR machine.

One of our aims is to test **new photosensitizers** and those coupled with carriers such as polymers and nanoparticles, or such as proteins, peptides and antibodies to improve solubility and targeting function of the photosensitizers. Another aim is to investigate **signaling pathways leading to cell death** (e.g. apoptosis), **adaptive response or survival** following PDT. Furthermore we try to analyze the **immunogenicity** observed following PDT. Based on a mouse study, where we showed complete tumor eradication und immunity following low dose PDT, we search for factors responsible for immune stimulation, e.g. damage-associated molecular patterns or shifts in the T-cell subsets.

Another focus is on the cellular and molecular basics of the **health effects of low-level-alpha-radiation** of radon spas.

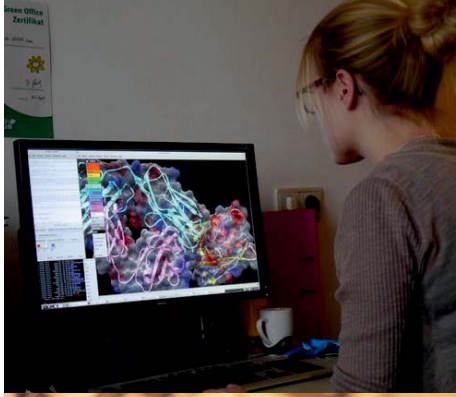
We cooperate with groups from our faculty, the PMU and SALK as wells as with national and international researchers and with the industry (Sanochemia, Tecan, Planta, W&H).

Team Peter Lackner
Division of Allergy and Immunology

Our team concentrates on the development and a**pplication of bioinformatics methods in the area of immunology and allergy research**. The objects of our investigations are proteins. These molecules are major players in immunological pathways but also origin of allergies. In particular we are interested in allergens. We aim to contribute to answer two central questions: **Why are some proteins allergens while closely related ones are not?** And, how can we transform natural allergens by protein engineering into drugs (hypoallergens) for allergy treatment.

To come closer to answers, we **analyze, compare and predict properties of these proteins like the 3D-structure** per se, molecular functions, interactions, stability or flexibility. Subsequently we try to modify certain properties of the allergens to deliberately change their molecular behavior and make them accessible as drugs for allergy treatment. Our **research is highly interdisciplinary** and is performed in close collaboration with experimental groups of the Division of Allergy and Immunology. In terms of computational problems we collaborate with the Universities of Applied Sciences in Salzburg and Upper Austria.

For the analysis and prediction tasks we use a panel of publicly available tools such as **molecular graphics, molecular modeling, sequence and structure comparison**, etc. Occasionally, the publicly available methods are not tailored for specific tasks or they are simply insufficient in terms of reliability or performance. Other methods are circuitous to use, especially when applied to larger data sets. In these cases we implement our **own software solutions to overcome the deficiencies using modern technical approaches like machine learning or distributed computing**. Bioinformatics methods developed in our team are often of broader interest and thus they are released to the community.



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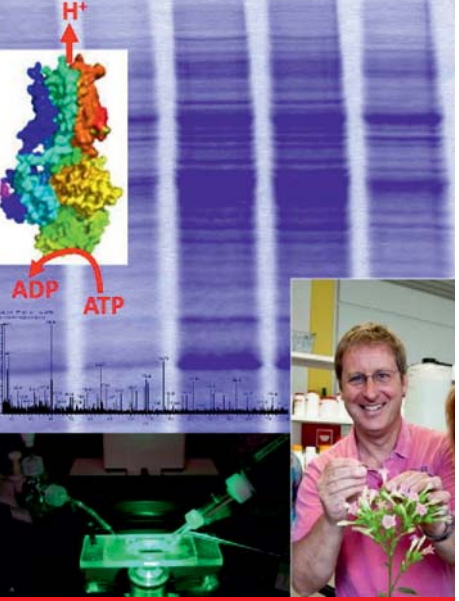

Team Gerhard Obermeyer
Division of Molecular Plant Biophysics and Biochemistry

The research focusses on the central biological question how cells develop and maintain **polarity**. An extreme example of polarity is the growth of **pollen** tubes: the growth zone is restricted to the first 20 µm of the tube tip but pollen tubes can reach a length of several centimeters while maintaining a diameter of only 10 µm. Furthermore, growth of pollen tubes through the style tissue is a prerequisite for successful fertilization which guarantees high **crop yields** for human nutrition. Future problems caused by **global warming** are already addressed in today's research and the effects of high temperature and draught on pollen fertility are studied, too.

To understand the molecular mechanisms behind pollen tube growth, the function, structure and activity of **membrane proteins** is studied using a broad panel of different methods ranging from single molecule to systems biology approaches: **biophysical techniques** like patch-clamp, turgor pressure and ion-sensitive electrodes, microscopy methods to localize membrane proteins, protein interactions or cytosolic ion concentrations in living cells by fluorescence resonance energy transfer or ratio imaging using fluorescence tagged proteins or genetically engineered **nanosensors**, respectively, and biochemical methods as well as up-to-date **omics techniques** (proteomics, metabolomics, transcriptomics) to reveal functional **protein complexes** in the plasma membrane and active **signaling pathways**. Recent publications include the characterization of a glutamate receptor-like protein in plants (Michard et al 2011, Science 332: 434) and the world-wide first study on the pollen metabolome (Obermeyer et al 2013, Plant Physiology 1822).

The group is well-established in an international and national research network: students and co-workers from all over the world (Australia, Czech Republic, Egypt, France, Germany, Greece, India, Israel, Italy, Mexico, Myanmar, Spain, Turkey, UK, USA) have already joined the group. We welcome students for Bachelor-, Master and doctoral thesis who are interested in working on innovative aspects of plant sciences in a stimulating and pleasant atmosphere.

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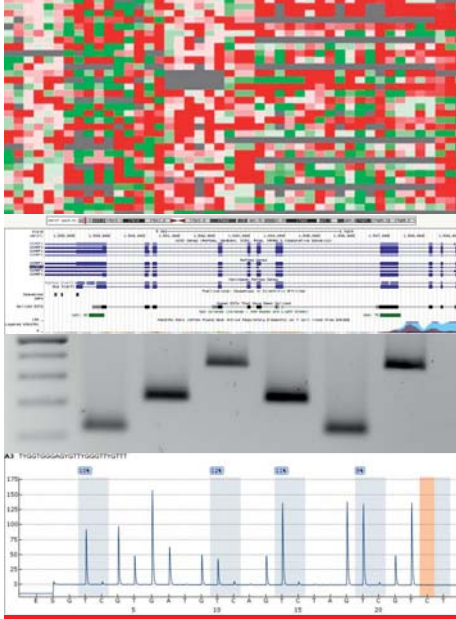
Team Angela Risch
Division of Molecular Tumor Biology

The research of our team focuses on **genetic and epigenetic variations and aberrations in the context of cancer**. Interindividual genetic and epigenetic variation, as well as acquired genetic and epigenetic changes can affect cancer risk, tumor development and clinical prognosis. Methylation patterns differ across tissues, but can also change as a result of age, disease or exposure, e.g. to tobacco smoke. We are interested in their potential **use as biomarkers of exposure, or as diagnostic/prognostic markers**. Epigenetic dysregulation may also point us to important new mechanisms in carcinogenesis.

We employ a broad spectrum of molecular biological techniques with appropriate bioinformatic and statistical analysis. Most importantly, we employ a range of methods for **methylome analysis**, e.g. in clinical samples, and then use sequence specific quantitative methylation analysis to validate findings and to better characterize the epigenetic dysregulation. This is further followed by in vitro analyses of target genes or miRNAs for functional characterization.

Within large international consortia we investigate **single nucleotide polymorphisms (SNP)** and their functional consequences as risk factors for disease, with a particular emphasis on lung cancer. Within **genome-wide SNP association studies**, risk regions have been defined, but the mechanisms of such associations mostly remain unclear. We are now determining methylation patterns at high resolution, and are looking to correlate SNPs with epigenetic patterns with the aim of identifying functionally relevant SNPs and mechanisms promoting carcinogenesis.

Inflammation-related epigenetic changes and alterations in epigenetic patterns as a result of tumor-microenvironment interactions hold particular promise in the context of identifying potentially **clinically useful biomarkers. Epigenetic drugs** are being used in the context of cancer treatment, but much remains to be learned about their mechanisms of action and optimal use.



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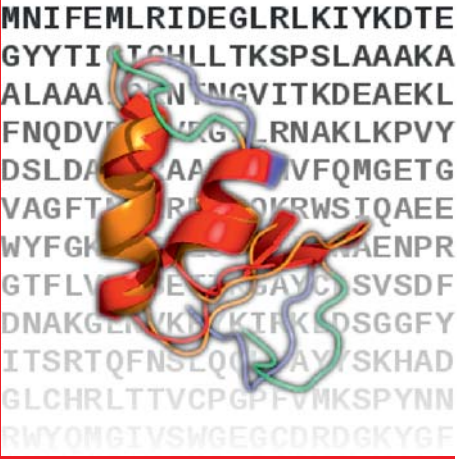
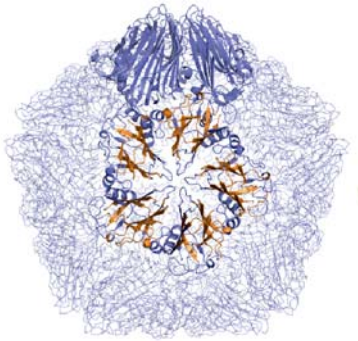
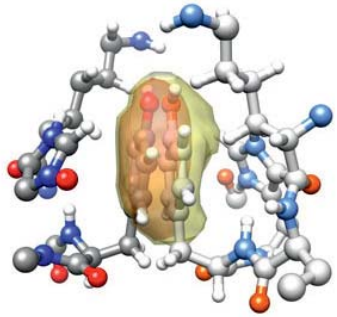


Team Manfred Sippl / Markus Wiederstein
Division of Structural Biology & Bioinformatics

Our major research programmes concern the **validation, refinement, prediction, and classification of protein structures**. We develop **computational tools** and programs for protein structure comparison, high speed structure data base searches and protein domain decomposition. We use **statistical thermodynamics and information theory** to derive energy functions for protein structure validation, refinement and prediction. We employ Python and C/C++ for the implementation of computer source code. We share finished programs with the scientific and commercial communities.

Current major research projects include: comparison and alignment of **three-dimensional structures of proteins**; fast and sensitive **structure data base searches**; methods for the automated decomposition of protein structures into domains; understanding the **forces that govern protein folding and protein stability**.





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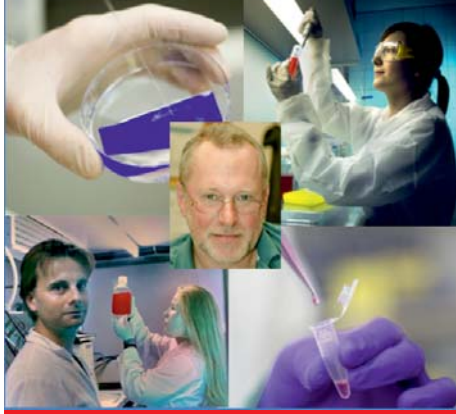
Team Josef Thalhamer / Richard Weiss
Division of Allergy and Immunology

Due to the increasing prevalence of type I allergy, there is an urgent need for novel therapeutic but also prophylactic approaches against this disease. We have long lasting experience in the field of intradermal **genetic immunization** and have demonstrated the proof of concept for tailor-made allergy vaccines based on plasmid DNA and messenger RNA. The optimal safety profile makes RNA a promising candidate for a first human prophylactic vaccine for early protection against allergic sensitization. While prophylaxis may be the most efficient intervention, the high incidence of allergic diseases calls for improved therapeutic vaccines. Allergen specific immunotherapy suffers from low patient compliance due to the need for a multitude of subcutaneous injections over several years and the risk of systemic side effects.

The skin has been rediscovered as an effective route for vaccination, not only for gene vaccines but also for protein vaccines. It is rich in antigen presenting cells and is efficiently drained via local lymph nodes. We have recently established **transcutaneous immunotherapy** via laser-generated micropores, which displays similar efficacy compared to subcutaneous immunization. Using glycoconjugates of allergens and carbohydrates, we design **nanoparticles with decreased allergenicity** (so-called hypoallergens) and increased immunogenicity, which specifically target skin dendritic cells via different receptors.

These translational research approaches are based on studies of **cellular and molecular mechanisms of skin immunity**, with a focus on immune polarization and allergic sensitization. Currently, we use state-of-the-art transgenic mouse models to investigate **immune functions of epidermal Langerhans cells** by inducible antigen expression after genetic immunization, and the influence of **structural stability of proteins** on immunogenicity and **immune response polarization** using in silico mutation and screening approaches followed by wet lab analysis.



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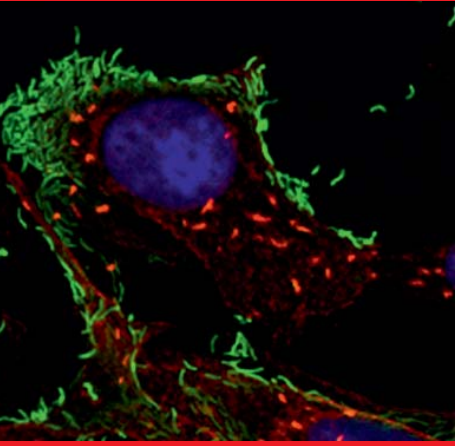

Team Silja Weßler
Division of Microbiology

Precise regulation of **signal transduction** is required to control normal biological processes. Pathogens developed fascinating strategies to deregulate cellular signaling pathways, which have been related to a number of disorders, ranging from relatively non-life-threatening disorders to extremely virulent diseases such as cancer as a consequence of growth control loss and resistance to apoptosis. Focus of our research is the investigation of the molecular mechanisms of how the bacterial **class-I carcinogen *Helicobacter pylori*** interferes with host cell functions leading to gastric carcinogenesis.

H. pylori induces depolarization and migration of epithelial cells, which is enhanced by translocation of the pathogenic factor CagA into host cells. Once injected into the cytosol CagA is rapidly phosphorylated by Src family kinases. We identified the non-receptor tyrosine kinase c-Abl as an additional crucial mediator of *H. pylori*-induced migration and novel CagA kinase, which maintains CagA phosphorylation in epithelial cells. As **Src and c-Abl kinases** are important in driving cells toward neoplastic transformation they represent a promising field in future treatments of gastric cancer progression. In current projects we investigate those derailed non-receptor tyrosine kinases in *H. pylori* associated carcinogenesis.

Depolarization of epithelial cells also implies the disruption of E-cadherin-mediated adhesion junctions (AJs). We analyze the disintegration of E-cadherin-dependent AJs and identified the **serine protease HtrA** as a new secreted virulence factor of *H. pylori* that directly cleaves the E-cadherin ectodomain leading to the disruption of the epithelial barrier functions and allow *H. pylori* to access the intercellular space. Since HtrA-mediated **E-cadherin** cleavage appears to a prevalent mechanism in bacterial infections we analyze the functional consequences of HtrA activity in the pathogenesis of a wide range of further gastrointestinal pathogens (e.g. *Campylobacter jejuni, or Listeria monocytogenses, etc*.) and develop **inhibitory compounds** to prevent HtrA-dependent pathogenesis.

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Team Peter Hammerl: the Central Animal Laboratories

Owing to the well-established methodology for genetic manipulation, **laboratory rodents** have become key tools for the investigation of **gene function in physiologic and pathologic settings**. Despite all apparent differences, their close genetic relationship to humans makes genetically modified rodents highly relevant models for **biomedical research**. As a **service department** for the university's work groups the central animal laboratories maintain **more than 150 genetically manipulated mouse and rat strains,** mainly for research in immunology and tumor biology. The central animal laboratories are a **user institution authorized and registered**, and subject to annual audit, by the competent national authorities. The department comprises areas with conventional hygiene level, as well as **specified-pathogen-free (SPF) areas**. The SPF area includes a high-hygiene barrier for the breeding of particularly sensitive immune-deficient lines and an experimental area where researchers have direct access to their animals.

The entire facility is ventilated from its own air-condition unit, supplying **temperature and humidity-controlled HEPA-filtered air** to all rooms. A positive pressure cascade protects animal rooms from the entry air-borne pathogens. Personnel enter SPF-areas through air-controlled locks and wear protective gown, shoes, caps, masks and autoclaved overalls. In the SPF area, animals are accommodated in **individually ventilated cage (IVC) systems** that supply sterile-filtered air to the cages and filter exhaust air before being released into the central exhaust air collection system. All working procedures involving animals are carried out under sterile conditions, using **laminar flow work benches**. Staff is responsible for **general husbandry,** and supplies animals once a week with autoclaved **cages, bedding, nesting materials, food and drinking water.** The **embryo transfer unit** offers services such as rederivation of potentially contaminated mouse lines, cryo-conservation of sperm, or delivery of tissue biopsies. A rigorous **health monitoring program** is in force to screen animals, on a regular basis, for relevant pathogens. The **designated veterinarian** and the institution's **animal welfare body** support staff and researchers in all aspects of animal care, welfare and experimentation, and organize **specific education and training programs** in laboratory animal science, both for scientists as well as animal care technicians.

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