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sischen Krebstherapien eignen sich die genetisch modifizierten Mausmodelle auch, um die Entwicklung von neuen Therapiemöglichkeiten zu begleiten und zu steuern. Ein Beispiel ist die Entwicklung von PARP Inhibitoren, welche eine «synthetische Letalität» vom BRCA1/2-defizienten Tumoren verursachen. In den Mausmodellen für BRCA1/2-mutierten Brustkrebs konnten wir die Wirkung des PARP Inhibitors olaparib (Lynparza®) zeigen und haben einen Vorschlag für die klinische Anwendung in Kombination mit Platinmedikamenten gemacht. Der Nutzen von olaparib konnte dann auch in klinischen Studien bestätigt werden und kommt nun mit dem kürzlich durch die FDA und EMA zugelassenen Lynparza® zur Anwendung. In unseren Modellen haben wir dann auch Mechanismen der Therapieresistenz identifiziert, welche für PARP Inhibitoren von klinischer Bedeutung sein könnten. Neben dem aktiven Transport aus der Zelle durch P-gp/ABCB1 haben wir insbesondere Mechanismen gefunden, welche die DNA Reparatur zumindest teilweise wiederherstellen. Für diese Identifikation hat sich die Kombination von funktionalen in vitro Screens in 2D Kulturen mit der Analyse von resistenten Maustumoren als nützlich erwiesen. Wir konnten zeigen, dass durch den Verlust von 53BP1 oder REV7 resistente Zellen in der Lage sind, die HR auch ohne BRCA1 durchzuführen. Folglich sprechen sie nicht mehr auf die PARP Inhibitoren an. Des weiteren zeigen unsere Studien, dass es noch einige weitere Mechanismen der PARP Inhibitor-Resistenz geben muss, welche wir noch nicht verstehen. Insbesondere ist unklar, wie BRCA2-defiziente Tumore sich anpassen und dann die tödliche Wirkung der PARP Inhibitoren verhindern.

Eine weitere interessante Beobachtung in unseren Tiermodellen ist die Antwort auf Platinmedikamente wie cisplatin und carboplatin. Im Gegensatz zu anderen Therapien kommt es hier nicht zu einer sekundären Resistenz. Dennoch werden nicht alle Tumorzellen durch die Therapie vernichtet. Von wenigen übrig gebliebenen Tumorzellen wachsen wieder neue Tumoren aus, welche dann genauso auf die Therapie ansprechen wie die Therapie-naiven Tumore. In unserem BRCA1 Model haben wir keine Anzeichen gefunden, dass Krebsstammzellen spezielle Abwehrmechanismen gegen die Therapie haben. Stattdessen haben wir Hinweise, dass die überlebenden Zellen in einen Schlafzustand gehen, und nach Beendigung der Therapie wieder erwachen. Die genauen zugrunde liegenden molekularen Mechanismen können wir nun genauer studieren. Insbesondere hoffen wir, in unseren Modellen neue Therapiemöglichkeiten zu finden, welche die Schläferzellen komplett vernichten können.

Durch das Zusammenspiel von Grundlagenforschung an unterschiedlichen in vitro und in vivo Modellen mit der Untersuchung von Patientenmaterial haben wir eine wirkungsvolle Waffe, um die Hürde der Krebstherapieresistenz zu bekämpfen. Ein Punkt, der hier noch verbessert werden kann, ist die Validierung von grundlegenden Mechanismen, welche in den Modellsystemen gefunden werden, in Tumoren des Menschen. Hier denken wir, dass die vergleichende molekulare Pathologie gestärkt werden sollte. Um dieses Ziel zu erreichen, haben wir in Zusammenarbeit mit der Humanpathologie in Bern die COMPATH Plattform aufgebaut. Wir hoffen, dass hiermit die wissenschaftliche Zusammenarbeit zwischen den grundlegenden Naturwissenschaften sowie der Human- und Veterinärmedizin gestärkt werden kann, um gemeinsam neue Strategien für die Bekämpfung von Krebserkrankungen zu erarbeiten. ■

The Institute of Veterinary Biochemistry and Molecular Biology
An example of how understanding of disease processes at the molecular level contributes to defining strategies for effective therapy and diagnosis

Michael Hottiger*

The Institute of Veterinary Biochemistry and Molecular Biology (IVBMB) is a preclinical Institute of the Vetsuisse Faculty at the University of Zurich (UZH) located at the Irchel Campus within the Science Faculty, to which it has a long tradition of strong collaborative interactions. The Institute is responsible for undergraduate teaching within the Vetsuisse-Faculty and makes important contributions to the post-graduated DVM and PhD education (as part of the Life Science Zurich Graduate School) at the UZH.

The IVBMB has a strong commitment to translational biomedical research, linking basic research to clinically relevant questions, thereby helping to close the gap between preclinical and clinical research. As such, it hosts the interfaculty Competence Center for Applied Biotechnology and Molecular Medicine

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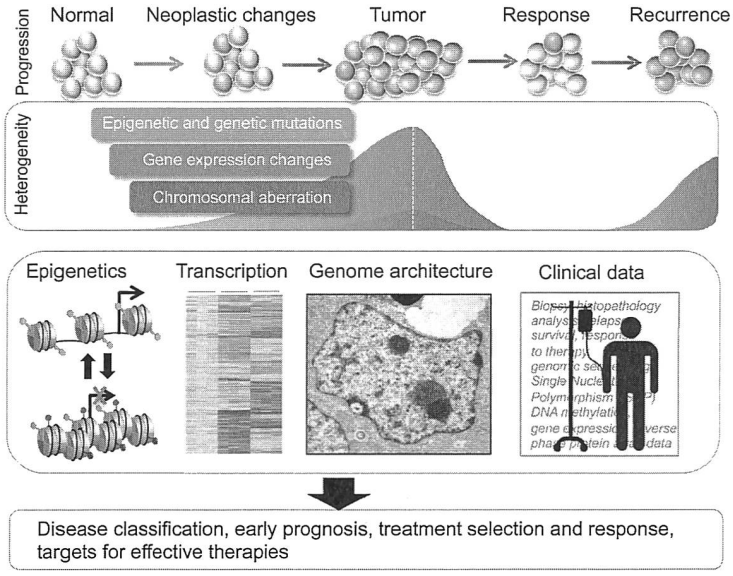

Figure 1 *(coulours see web version). Epigenetics and chromatin dynamics in cancer. As cancer progresses, mutations at the genome and epigenome accumulate, causing alteration of gene expression and rearrangements. Some of theses changes drive resistance to therapy and inevitable relapse and recurrence of the cancer. Our research aims to understand how epigenetic regulatory systems contribute to cancer initiation, progression and resistance to therapy by studying processes implicated in the regulation of transcription, genome architecture and inheritance of chromatin states in physiological and malignant conditions. Our research strategy is based on functional analyses using in vitro and in vivo models, bioinformatics approaches and clinical information that together will be instrumental to answer key questions such as disease classification, early prognosis, treatment selection and response, and disease progression.*

(CABMM) to foster the translation from basic research to clinical applications.

The mission of the IVBMB is to elucidate the molecular mechanisms leading to disease, to lay the foundation for novel therapeutic strategies and to offer a comprehensive and modern teaching program to students at different levels of biomedical education. The underlying vision is to be at the forefront of biomedical research and teaching, and to provide an internationally recognized center for the development of new solutions in translational biomedicine. Due to its interdisciplinary and translational research approach, the IVBMB is second to none within the Vetsuisse Faculty, has a unique position in Switzerland, and thus is of exceptional national importance with an excellent international reputation and impressive research output in leading scientific journals.

Currently, the IVBMB has four main research lines, organized in four independent research groups, which are entirely funded by competitive research grants (e.g. SNF grants). Their research is centered on different chromatin signaling pathways that regulate the response to pathogens or cellular stresses (see individual project descriptions below).

1. Epigenetics and chromatin dynamics in cancer (Santoro group)

Despite sharing the same genome, different cell types respond differently to environmental, developmental or metabolic cues. This variable property is a defining aspect of a cell's identity and is mainly interpreted at the level of epigenetic signature and chromatin organization. Perturbations of this signature or organization can lead to congenital disorders or predispose people to acquire disease states such as cancer.

The aim of our research is to understand how epigenetic regulatory systems contribute to the susceptibility and development of complex diseases. We do this by studying the molecular processes implicated in cellular memory, cell fate specification and cancer initiation-progression. Our previous work has offered important examples of epigenetic regulation and underscored the role of long non-coding (lnc)RNAs in the epigenetic network that ultimately defines gene expression programs.

Our recent work has shown that lncRNA processing and the cell nucleolus are active regulators of chromatin plasticity, challenging current views on heterochromatin regulation and function in cell pluripotency and lineage commitment. Given the growing connection between epigenetic abnormalities in cancer and stem cell-like traits, these results provide important information on self-renewal and pluripotency states, features that characterize the most aggressive cancers. Moreover, our work has highlighted the role of epigenetics in prostate cancer, the most common non-cutaneous malignancy in men. The results have provided evidences that the epigenetic factor TIP5 is involved in prostate cancer progression and is a potential early prognostic biomarker to identify aggressiveness in patients diagnosed of indolent cancer, linking molecular findings with recurrence in clinical samples. These results are not only instrumental to predict metastatic potential at an early stage but also offer new therapeutic strategies to target the critical cell population resistant to conventional treatment regimens, such as androgen ablation therapy and/or treatment with androgen receptor antagonists.

In summary, our research strategy is based on functional analyses using in vitro and in vivo models, bioinformatics approaches and clinical information that together will be instrumental to answer key questions such as disease classification, prognosis, treatment selection and response, and disease progression (Fig. 1).

Further information on the research carried out by

the Santoro group can be obtained at: http://www.vetbio.uzh.ch/Research/DrSantoro.html

2. DNA and chromatin integrity in developmental disorders (van Loon group)

Accumulation of DNA lesions influences aging, development of cancer and neurological disorders. Base excision repair (BER) removes damaged DNA bases and maintains genetic integrity (Fig. 2). While different steps of this repair reaction are known, it is unclear how BER occurs within chromatin. Also, though many human diseases correlate with accumulation of damaged bases, data documenting the possible contribution of BER glycosylases and DNA polymerases (Pols) to pathological conditions is limited.

We aim to explore how impaired BER contributes to the onset of human pathologies by focusing on (i) the influence of chromatin organization on repair efficiency, (ii) identification of factors that influence repair of damaged bases, and (iii) the posttranslational mechanisms that regulate the levels of BER enzymes and modulate repair capacity.

Recently, we have demonstrated that chromatin organisation negatively influences BER activity and that the presence of chromatin modifiers is a prerequisit for efficient repair, as well as the maintanance of genome stability. Besides chromatin, posttranslational mechanisms affect repair capacity. Mutations in E3 ubiquitin ligase are associated with the human disease X-linked intellectual disability (XLID). The underlying pathomechanism of this neurodevelopmental condition is largly unknown. Very recently, we have provided evidence that inefficient BER of oxidative DNA damage and subsequent accumulation of mutations could drive E3 ubiquitin ligase-associated XLID.

Increased levels of ribonucleotides and oxidative DNA base lesions are tightly associated with neurodegeneration. Novel findings by us indicate that BER Pols abundantly expressed in adult neurons incorporate significant amounts of ribonucleotides opposite oxidative DNA lesions and thus could contribute to neurodegeneration. These results have the potential to not only provide insight into the pathological aspects underlying neurological disorders, but also of diseases such as cancer.
Further information on the research carried out by the van Loon group can be obtained at: http://www.vetbio.uzh.ch/Research/DrVanLoon.html

3. Genome instability in cancer and aging (Altmeyer group)

Human cells contain about six billion building blocks that together constitute our genetic material, the

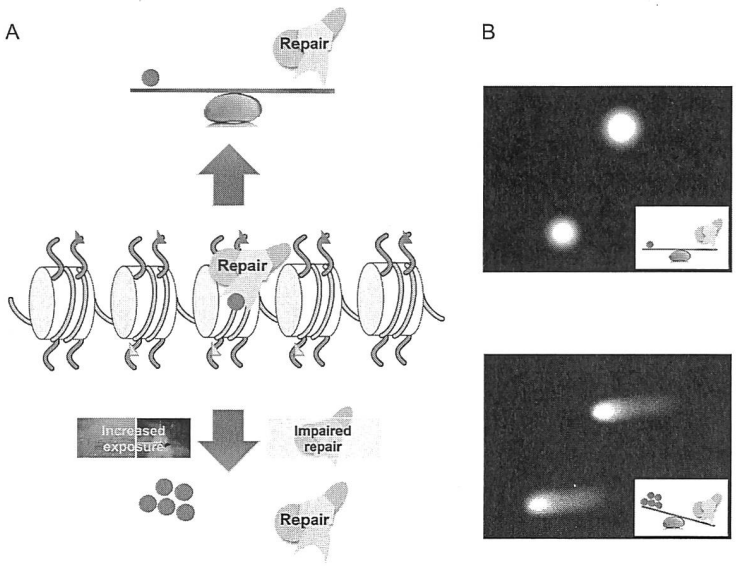

Figure 2 *(coulours see web version). DNA and chromatin integrity. (A) Lesions (red circles) are generated every day in DNA. To maintain genome integrity, repair pathways have to act in the context of chromatin and remove DNA lesions. If the amount of lesions exceeds the repair capacity due to increased exposure to damaging agents or impaired DNA repair, lesions will accumulate leading to genome instability and potentially onset of different pathologies. (B) We employ a wide range of techniques allowing to analyse the impact of chromatin on the amounts of DNA lesions, as well as to compare DNA repair capacities between cells of healthy individuals and patients suffering from developmental disorders. Presented is a result of one such method, FLARE - single cell gel electrophoresis assay, revealing the increased amount of DNA lesions in patient cells (lower panel) compared to cells from a healthy individual (upper panel).*

DNA. The slightest damage to a single DNA base pair or alterations of the DNA sequence can lead to dysfunctional gene products, which in turn can greatly affect cell function and cause disease. To avoid such detrimental changes to our genetic material, cells have developed sophisticated molecular mechanisms to minimize DNA damage and efficiently repair genetic lesions when they occur. How cells coordinate damage-induced transactions in space and time and how enzymatic reactions are being kept in check to ensure faithful DNA replication and gene expression programs to operate without causing genomic instability remain pivotal, largely unresolved questions.

Our research aims at elucidating cellular mechanisms of genome integrity maintenance. Many of these mechanisms operate at the level of chromatin, and we investigate how different chromatin states affect DNA repair reactions, and how the DNA repair machinery itself uses spatially and temporally confined chromatin modifications to safeguard genome integrity. Such modifications can transiently subdivide the intracellular space by generating dedicated repair compartments surrounding DNA break sites. Our group thus studies their physicochemical properties and their functional contributions for

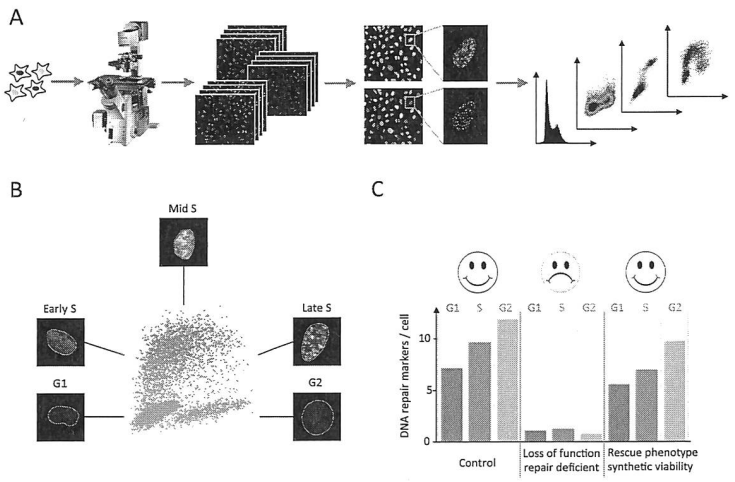

Figure 3 *(coulours see web version). Cell biology of genome integrity maintenance. (A) The Altmeyer group employs a dedicated high-content microscopy pipeline combined with software-assisted cell segmentation and quantitative feature extraction to interrogate cellular responses to genotoxic stress. Using this experimental setup, we are developing novel read-outs that allow us to combine cell proliferation data with information on sub-cellular structures such as the compartments generated transiently by cells to repair DNA lesions. (B) Example of how we use high-content microscopy data of cell populations to define the different stages of the cell cycle and investigate cellular features of genome integrity maintenance with high spatial and temporal resolution. (C) One of the current projects aims at identifying rescue phenotypes of DNA repair malfunction, in the hope to reveal concealed molecular interactions with potential implications for personalized cancer therapy.*

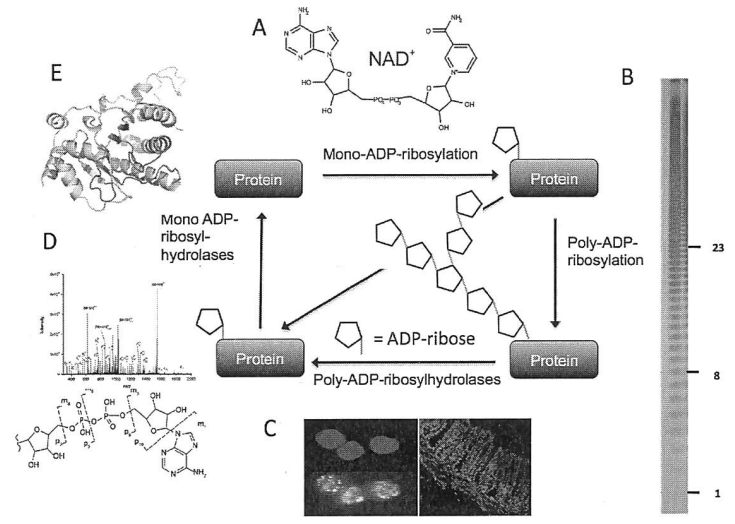

Figure 4 *(coulours see web version). Toward understanding the biomedical relevance of inflammation-induced ADP-ribosylation. Protein ADP-ribosylation is a reversible posttranslational modification (PTM) that can be investigated at different levels. The ADP-ribosylation cycle (center image) starts with mono-ADP-ribosylation, which can be subsequently extended to a poly-ADP-ribose. (A) NAD+ is the substrate for ADP-ribosylation, (B) polymers of ADP-ribose can be isolated and separated on a gel or (C) visualized by immunofluorescence in cells treated with hydrogen peroxide (left) or in tissues (right). Nuclei (in blue) were visualized by staining with DAPI. (D) Modified proteins and ADP-ribose acceptor sites can be identified by mass spectrometry. (E) Structure of the Macrodomain 2 (model based on the PDB ID: 2X47 crystal structure of MDO1), an enzyme able to hydrolyze mono-ADP-ribose attached to proteins. The primary macrodomain binding site loops are marked in red.*

chromatin transactions in the course of the DNA damage response. Moreover, we are interested in the principles that prevent DNA damage-induced chromatin modifications from turning into derailed reactions, which bear the risk to unbalance repair pathway choices and thereby undermine genome integrity. To achieve these research aims, we use state-of-the-art cell biological tools combined with molecular biology and biochemical approaches (Fig. 3). A major focus lies on quantitative high-content multivariate imaging of cell populations exposed to genotoxic stress treatments. By combining this tailored cell imaging setup with targeted perturbations of cell functions through chemical and reverse genetics, our group explores hitherto unknown genome caretakers and their modes of action.

The mechanisms that safeguard genome stability are often subverted in cancer and in certain age-related disorders. A detailed molecular understanding of the cellular pathways that maintain genome integrity and how they are deregulated in human disease is essential to comprehend when and why major cellular functions start to fail. Beyond a potential for early detection and diagnosis, research dedicated to elucidate principles of chromatin and genome biology has also direct therapeutical implications: novel approaches for cancer treatment based on synthetic lethal interactions of cancer therapeutics with cancer-specific DNA repair defects are currently being tested in advanced clinical trials, and our group efforts aim at contributing to this promising avenue of targeted cancer therapy.

Further information on the research carried out by the Altmeyer group can be obtained at: http://www.vetbio.uzh.ch/Research/ProfAltmeyer.html

4. Cell signaling in inflammatory diseases (Hottiger group)

Inflammation is a complex reaction of cells in response to pathogens, cell damage or harmful molecules with the intended purpose to regenerate damaged tissues. Dysregulation of the inflammatory response results in many adverse medical conditions, such as cardiovascular diseases or the development of cancer. One of the most important regulators of the host inflammatory response is a factor called NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells), which activates the transcription of genes that encode specific inflammatory signalling molecules. During the last years, it has become clear that the function of NF-κB function is orchestrated by the chemical modification of proteins that regulate the chromatin structure. One such chemical chromatin modification is ADP-ribosylation, which is one of the key regulatory elements in the control of

inflammation and a possible target for pharmacotherapy.

The aim of our research is to understand the molecular signaling processes and mechanisms that regulate chromatin changes and NF-κB-dependent gene expression through protein ADP-ribosylation.

Over the last decade, we have significantly contributed to fundamental insights into the function of ADP-ribosylation during the innate immune response. We were the first to show that ARTD1 regulates NF-κB-dependent gene expression. At the molecular level, we have shown that non-apoptotic LPS-induced caspase 7 activation via the NLRP3 inflammasome induces ARTD1 cleavage at the transcriptional start site of distinct NF-κB target genes and thereby causes elevated expression of these genes. For ADP-ribosylation to act as a regulator, the modification must also be removed once the signalling function has been fulfilled (Fig. 4). We have identified macrodomain-containing proteins (e.g., MacroD1 and MacroD2 for aspartic and glutamic acid) to be able to carry out this activity (i.e. to function as eraser). In addition, our research group is at the forefront of developing mass spectrometry approaches, which for the first time has allowed to identify all ADP-ribosylated proteins in cells, to define ADP-ribosyl modifications as specific markers for stress conditions and to study the sensitivity of inflammatory and cancer cells to clinically used ADP-ribosylation inhibitors. Furthermore, we have also provided evidence that inhibitors of ADP-ribosylation significantly reduce *Helicobacter*-induced neoplasia or the generation of atherosclerotic plaques.

Understanding the regulatory mechanism of NF-κB-dependent gene expression by ADP-ribosylation will not only improve our understanding of basic biochemical reactions and physiological processes, but may generate new insights concerning the onset and development of inflammation and inflammation-associated diseases and contribute to the translation of scientific knowledge into clinical medicine.
Further information on the research carried out by the Hottiger group can be obtained at: http://www.vetbio.uzh.ch/Research/ProfHottiger.html

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Veterinärdermatologie – Ein Beispiel Translationaler Medizin oder « from bench to bedside »

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1. Einleitung

Als übertierartliche Disziplin beschäftigt sich die klinische Veterinärdermatologie primär mit Problemen der Haut bei Hund, Katze und Pferd. Seltener sind unsere Patienten Kühe, kleine Wiederkäuer, Neuweltkameliden oder Meerschweinchen. Hautveränderungen sind ein häufiges Problem in der industrialisierten Welt und machen im Schnitt 20% aller Fälle in der Kleintierpraxis aus.

Durch ein internationales Ausbildungsprogram in Veterinärdermatologie und der globalen Vernetzung der Hautspezialisten von Europa über Amerika bis Japan hat sich die Disziplin der Veterinärdermatolo-