Research Programme of the Paul-Ehrlich-Institut 2016 – 2020

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1. Preamble

The combination of regulatory and research tasks at the Paul-Ehrlich-Institut (PEI) is unique among the European medicines agencies. Due to this sui generis position, we are especially qualified to provide science-based expert advice on questions concerning the quality, efficacy, and safety of vaccines and biomedicines. In this spirit, the present research program presents the areas and goals of our research activities. Our strategic vision is to set new standards in the field of vaccines and biomedicines for the protection of both human and animal health as an internationally leading regulatory authority and a competitive life science research institute. In this regard, our research program shall give a clear overview on the amalgamation of regulatory and research duties, define an appropriate general framework for expedient research activities and present a concise vision on the future research strategy to meet emerging PEI-specific challenges. More detailed information on research activities in the various research groups[1] and their cooperation within national and international research consortia[2] can be found on our website. In addition, our bi-annual report provides an excellent overview on regulation and research at the PEI for the general public[3].

[1] http://www.pei.de/research-groups
[2] http://www.pei.de/research-programme
[3] http://www.pei.de/annual-reports

2. Introduction

The PEI is the German Federal Institute for Vaccines and Biomedicines reporting to the Federal Ministry of Health (Bundesministerium für Gesundheit). While vaccines are defined as substances that are administered to humans and animals with the aim to induce immunological protection against a particular disease, the term biomedicine comprises therapeutic medicinal products containing or derived from biological substances (biologicals).

The "Gesetz über die Errichtung eines Bundesamtes für Sera und Impfstoffe" (act of July 7, 1972)[4] assigns regulatory and research tasks to the PEI. Today, the institute's regulatory activities relate to various responsibilities laid down in German and European legislation. We are in charge of clinical trial approvals and responsible for the benefit-risk assessment and marketing authorization of vaccines for human and veterinary use as well as biomedicines for human use. The latter includes antibodies, allergens, gene and (stem) cell therapy products, tissue engineered products, and blood and plasma-derived products. In this context, we also record and analyse notifications of observed or suspected adverse reactions. This constitutes the basis for the identification of risks associated with a product, and for the coordination of measures to prevent direct or indirect hazards to human or animal health (pharmacovigilance). These tasks are complemented by inspection activities and the official batch release testing of vaccines and biomedicines prior to marketing.

In accordance with our legal responsibilities and the "Konzept einer modernen Ressortforschung"[5] outlined by the German Federal Government, the institute has defined its mission as promoting and safeguarding the quality, efficacy, and safety of vaccines and biomedicines exploiting the synergism of regulation and research. Our structure, which extends the concept of the U.S. Food and Drug Administration (FDA) as a combined regulatory authority and research institute to an integrated "One Health"[6] approach by the conjoint responsibility for human and veterinary products, sets a unique high-level standard for other regulatory agencies in Europe and world-wide. It enables us to chaperon vaccines and biomedicines from development and marketing authorization all the way to the assessment of undesirable effects and the post-approval monitoring of quality, safety, and efficacy. As repeatedly emphasized by the German Science and Humanities Council, it is this combination of regulatory and

[4] http://www.gesetze-im-internet.de/bundesrecht/basig/gesamt.pdf
[5] https://www.bmbf.de/files/konzept_ressortforschung.pdf

research expertise that constitutes our international reputation and state-of-the-art competence[7].

Mandate of research at the PEI is to complement our regulatory responsibilities by providing solutions for innovative product testing and investigating questions relevant for our current and future regulatory decision making. In this spirit, we are committed to the field of regulatory science, which "constitutes the foundation of regulatory decision-making" and involves "basic and applied biomedical sciences, clinical trial methodology, and epidemiology"[8]. Our complementary concept of prospective basic research focusses on the understanding and development of innovative prophylactic and therapeutic strategies with an emphasis on novel vaccination routes and adjuvants, the efficacy of specific immunotherapy, the potential of targeted gene and (stem) cell therapy, and the treatment of cancer by oncolytic viruses and immunotherapeutics. Fulfilment of these statutory research tasks comes to the fore by internationally recognized publications in the fields of immunology, virology, microbiology, allergology, haematology, gene and cell therapy as well as pharmacoepidemiology.

One prerequisite for success in research and regulation is our collaboration with multiple national and international research institutions, regulatory authorities, and public health organisations. This includes our cooperation with the German Centres for Health Research[9], particularly the German Centre for Infectious Disease Research (DZIF) and the German Consortium for Cancer Research (DKTK), and our work as experts for the European Medicines Agency (EMA), the European Directorate for the Quality of Medicines (EDQM) and the World Health Organization (WHO). With respect to the development of innovative solutions for experimental product testing, the PEI is part of the Official Medicines Control Laboratories (OMCLs) network supporting regulatory authorities in controlling the quality of European-wide marketed vaccines and biomedicines. Moreover, the PEI is designated as a WHO Collaboration Centre for Blood Products and in vitro Diagnostic Devices and as a WHO Collaboration Centre for the Standardization and Regulatory Evaluation of Vaccines. One task is to support the WHO in the development of international standards and standardized assays for the experimental testing of the quality, safety, and efficacy of blood products and vaccines.

[6] https://www.bmbf.de/files/Forschungsvereinbarung_Zoonosen.pdf
[7] http://www.wissenschaftsrat.de/download/archiv/9860-10.pdf
[8] Elmgren, L., et al. (2013) Vaccine, 31S: B163-B175
[9] http://www.bmbf.de/de/gesundheitszentren.php

Our research projects provide important contributions to our state-of-the-art expertise for top-quality assessment of new biomedical strategies and innovative experimental approaches. Our research competence enables us to promptly take appropriate countermeasures in case of serious quality, safety and efficacy issues. To keep and promote this position, our research projects are designed:

- to ensure and improve the quality, safety, and efficacy of vaccines and biomedicines
- to foster the development of novel vaccines and biomedicines
- to innovate risk assessment in regulation
- to support evidence-based regulatory decisions
- to provide health policy decision-makers with expert advice regarding vaccines and biomedicines, and
- to further - where reasonable - the refinement, reduction and/or replacement of animal experiments in the field of product testing and batch release.

There are a total of 10 divisions in our institute, eight of which are responsible for complementary research and regulatory tasks in virology, microbiology, immunology, allergology, haematology and transfusion medicine, as well as gene and cell therapy. Six divisions hold regulatory responsibilities for specific sets of vaccines and/or biomedicines for human use. The regulation of immunological products for animal use is assigned to a separate division. A comprehensive division is responsible for pharmacovigilance assessments and appropriate corrective measures.

The heads of these divisions lead research groups focussing on projects to foster innovations in the fields of their specific responsibilities and, thus, to further state-of-the-art regulatory decisions, especially with respect to novel classes of vaccines and biomedicines. In addition, we have installed research groups of the president and vice president as well as independent junior research groups focussing on new and rapidly developing areas in the fields of our responsibilities.

To further exploit the existing synergy between regulatory and research activities and to facilitate interdisciplinary interaction across divisions and research groups, we have defined three key cross-divisional research areas:

- Regulatory Research & Innovative Medicinal Product Testing;
- Pathogen-Host & Biomedicine-Organism Interactions; and
- Experimental Vaccines, Therapies & Diagnostics.

These three key cross-divisional research areas constitute an integrative framework for the individual research foci of the divisions and research groups.

3. Cross-Divisional Research Areas

3.1. Regulatory Research & Innovative Medicinal Product Testing

Due to their biological origin, vaccines and biomedicines are far more complex than chemical-based drugs, and their composition may vary by batch. To account for this inherent variability, the PEI develops new experimental product testing approaches and standardized evaluation criteria, thus assuring product quality, safety, and efficacy. In addition, we investigate the cause of unexpected severe side effects associated with vaccines and biomedicines to provide an experimentally verified basis for our regulatory advice and decisions. To identify unmet needs for regulatory acceptance in current and past clinical development strategies, we systematically analyze reasons and rationales underlying the respective regulatory decisions. The resulting publications aid our international stakeholders in improving future applications for the approval of clinical studies or the marketing authorization of vaccines and biomedicines. In addition, we investigate how changes in national and international regulatory procedures impact patients and the medical and scientific communities.

3.2. Pathogen-Host & Biomedicine-Organism Interactions

The interplay between pathogens and the host immune system shares many similarities with biomedicine-organism interactions. The characterization of pathogen-host interactions constitutes the basis for a better understanding and the development of innovative prophylactic and therapeutic biomedicinal approaches against infectious diseases as well as non-communicable diseases such as cancer and allergies. At the same time, a detailed analysis of interactions between a biomedicine and its target organism is a prerequisite for both understanding the mechanism of biomedicine action and identifying the molecular cause of severe adverse effects. Research exploring these interactions is thus essential to maintain and improve the quality, safety, and efficacy of existing as well as novel biomedicinal approaches.

3.3. Experimental Vaccines, Therapies & Diagnostics

Building on insights from the other two research areas, the PEI explores innovative prophylactic, therapeutic, and diagnostic approaches. The respective research projects constitute the basis for our state-of-the-art regulatory competence regarding novel vaccines and biomedicinal products for human and veterinary medicine. We thereby not only foster innovation in medicine but also build the necessary knowledge base for regulatory decisions regarding these completely new product groups. One focus lies in the targeted modulation of immune responses, which plays an important role in the development of new vaccination strategies, specific immunotherapy of allergies, and regenerative medicine and cancer treatment approaches. In addition, we construct vector systems for the safe, efficient, and directed modification of target cells in gene and cellular therapy, and design oncolytic viruses to specifically eliminate tumor cells. These efforts are complemented by the identification and characterization of genetic or protein markers for the development of specific and sensitive diagnostics to advance the burgeoning field of personalized medicine.

4. Research Foci of the Divisions & Independent Research Groups

4.1. Division of EU Cooperation/Microbiology – Research for the Prevention & Therapy of Bacterial Diseases[10]

Head: PD Dr. Isabelle Bekeredjian-Ding

In respect to marketing authorization and batch release testing, the division is in charge of bacterial and parasitic vaccines and microbial safety of biomedicines. The biological origin of biomedicines poses the risk of bacterial and fungal contamination. Hence, methods for detection, elimination and inactivation of microbes are a prerequisite for licensing of biomedicines. In the clinic, the increasing multi-drug resistance of bacteria jeopardizes successful treatment of bacterial infections. Vaccines stimulating the immune system to specifically recognize and fight spread of multi-drug resistance strains could remedy this situation.

To advance microbial safety testing of biomedicines, we develop innovative assay systems enabling sensitive detection of harmful microbial bioburden including highly

transmissive resistance genes or microbial molecules provoking adverse immunological effects. Within the scope of our OMCL tasks, we develop standards and assays for vaccine potency and safety testing and implement them into official batch release procedures. Our aim is to significantly reduce the number of animal experiments required to ensure batch-to-batch consistency and exclude incomplete inactivation of harmful vaccine components such as bacterial toxins. One further focus in regulatory research is our interdisciplinary health care approach to analyse whether and how the general practitioner and the patient are made aware of changes in regulatory procedures, e.g. pharmacovigilance (safety) reporting for specific vaccines.

Under healthy conditions, commensal bacteria colonize the bodies' skin and mucosa without negative effects. This symbiotic state of tolerance is maintained by a fine-tuned interaction between microbes and host preventing bacterial eradication and host inflammation. In patients subjected to immunosuppression, surgery and/or intensive care, specific commensals are able to overcome the protective host response and exert pathologic effects. These infections are complicated by the lack of therapeutic options available for treatment of multi-drug resistance pathogens, which has become an increasing clinical threat. Working on the interaction of commensals with human immune cells, we investigate, which changes in bacterial expression of pathogenicity factors and alterations of the hosts' immune response synergize to promote infection and immunopathology. We aim at understanding specific immunity and immune memory to commensals and elucidating how commensals succeed in overcoming the hosts' immune response. These studies will be exploited to develop tailored vaccination strategies that revert the tolerogenic immune response in infection as well as vaccines that permit strain-specific eradication of multi-drug resistance commensals.

4.2. Division of Virology – Research for the Prevention & Therapy of Viral Diseases[11]

Head: Prof. Dr. Eberhard Hildt

The division is responsible for the marketing authorization and official batch release testing of viral vaccines, the identification and characterization of emerging viruses, the assessment of the viral safety of biomedicines, and the regulation of nucleic acid amplification tests (NATs) to detect viruses in blood (products). Leitmotif of our

[10] http://www.pei.de/microbiology-research
[11] http://www.pei.de/virology-research

regulatory and research efforts is notion of prevention as the most effective and safe action to combat viral infections.

To prevent infectious virus transmission via biomedicines, we develop and evaluate virus inactivation and removal methods as well as highly sensitive virus detection assays. Pursuing a consistency approach for the innovation of vaccine assessment, we develop assays for valid detection of antigen content and characterize vaccine-related antigen-antibody interactions. Our aim is to enable correlation of antigen content, antigen conformation and antigenicity with protective efficacy, and hence improve the prediction of vaccine quality. To contribute to the safety of existing vaccines, we strive to identify possible molecular root causes of associated adverse effects with a special focus on novel adjuvants. Moreover, we support the development of suitable regulatory standards to facilitate the translation of innovative vaccine concepts and novel NATs into the clinic by scientifically analysing regulatory bottlenecks.

Our studies to characterize virus-host interactions triggering viral replication and pathogenesis or influencing viral escape strategies are prerequisites for the development of novel prophylactic or antiviral strategies. In this regard, we especially focus on infections caused by highly variable viruses with the capacity to hide from the immune system (such as HIV, HBV, HCV and influenza viruses). We investigate how various virus genotypes and subtypes differ in their capacity to deregulate signal cascades interfering with antigen processing and presentation by the immune system. Our goal is to determine if these disparities are responsible for differences in virus-associated immunomodulation and pathogenesis, or rates of chronic infection development and response to therapy. In addition, we characterize functional and structural determinants of viral entry and release, replication, and morphogenesis, with the aim to identify novel targets for antiviral therapy and prognostic markers for outcome of infection or response to therapy.

Investigating virus vector technologies and cell permeable virus-like particles as epitope carriers, we work on prototypical recombinant vaccines enabling a fast track development of safe vaccines against new emerging viruses (such as dengue, chikungunya or Ebola). This approach also promises improved efficacy for combatting highly variable viruses that require e.g. a genotype-wide response. Therefore, we target the induction of both an efficient B cell response (combat of pathogens by antibodies) via suitable composition of the antigen’s epitope structure and a T cell response (cellular combat of pathogens) via mediation of cell permeability and targeted antigen presentation by competent immune cells.

4.3. Division of Immunology – Research to Elucidate Immune-Evasion Strategies of Human Pathogens & Safety Aspects of Immunological Biomedicines[12]

Head: Prof. Dr. Ger van Zandbergen

The division is responsible for the marketing authorization and official batch release testing of therapeutic antibodies and antisera. Moreover, the division is responsible for the regulation of specific therapeutic vaccines (such as tumour vaccines) and manages a cross-divisional OMCL service laboratory for the (physico-)chemical and immunological safety testing of biomedicines. Our regulatory research efforts are based on the division's OMCL expertise in the field of Centrally Authorised Products (CAP), which are selected for sampling and testing through a risk-based approach. Our skills to genetically modify primary human immune cells are essential for understanding the pathogenesis of obligate intracellular pathogens such as Leishmania parasites and Chlamydia bacteria.

Biosimilar antibodies have now been marketed for the first time. One important question is whether the safety and efficacy of these follow-on versions is equivalent to already authorized innovator biomedicines. Establishing and developing a unique set of sensitive analytical methods for the comparison of biosimilars with the innovator product enables us to understand the clinical relevance of potential molecular differences. We also exploit our expertise in the field of human immune cells to experimentally evaluate regulatory problems working with models close to the molecular pattern of human disease. In this regard, we currently focus on prototypic adjuvants and chemical constituents with the aim to identify their specific immunological effects and characterize their safety profile.

Pathogens hiding in immune cells have evolved multiple strategies to circumvent detection and prevent elimination. To understand the underlying pathogen-host interactions, we study pathogen-containing compartments and their development in primary human immune cells. To this end, we apply state of the art live cell imaging of compartment development and investigate host cell proteins involved in the dynamics of cellular defence. Combining this approach with electron microscopy analysis of the primary human host cell ultrastructure will enable us to understand how intracellular pathogens restrain an effective immune response. Genetic modification of primary human host cells finally allows us to investigate and manipulate susceptibility and

[12] http://www.pei.de/immunology-research

resistance factors on a molecular level. Our ultimate goal is to identify cellular factors that actually are able to eliminate intracellular pathogens. In collaboration with the University Medical Center Mainz, we also assess underlying mechanisms of an inefficient immune response in both tumour and infectious diseases. In this regard, we are especially interested in exploring the role of autophagy for the development of inefficient immunity. Moreover, we strive to identify inactivating factors of the immune system to target them for therapy.

Together, the research projects of our division make us experts in the fields of cellular and molecular immunobiology as well as immunological safety of biomedicines, supporting the quality of our regulatory decisions.

4.4. Division of Veterinary Medicine – Improving Animal & Human Health Through Better Understanding of Zoonotic Infectious Diseases[13]

Head: Dr. Veronika von Messling

The division is responsible for the authorization and experimental product testing of veterinary vaccines, sera, and novel immunotherapeutics. Our research focus complements this task by developing new prophylactic and therapeutic approaches against zoonotic or economically important diseases in the spirit of the “One Health Concept”, and by devising innovative strategies for in vitro product testing.

Our regulatory research efforts aim at improving product quality and safety with a special emphasis on refining and reducing animal experimentation in experimental product testing. We are currently developing new experimental approaches for the in vivo batch release testing of botulism toxin, rabies and avian encephalomyelitis vaccines with the goal of achieving official acceptance in the European Pharmacopoeia. Taking advantage of our in-depth understanding of veterinary immunological product development, we will continue to strengthen our leadership role in improving experimental medicinal product testing, giving new impulses to reduce the numbers of animals needed.

As most emerging and many well-known human pathogens are either transmitted from an animal reservoir or are closely related to animal pathogens, animal models provide unique insights in pathogen-host interactions. Focusing on influenza and morbilliviruses, we combine genetic manipulations of the respective pathogen with a

[13] http://www.pei.de/veterinary-medicine-research

detailed analysis of the resulting pathological and immunological changes in different animal models to identify immune correlates of protection and virulence factors. In the coming years, our emphasis will be on characterizing the role of the immune response in morbillivirus neuro-invasion and the impact of genetic diversity on tissue tropism.

With the increasing understanding of effective and aberrant immune responses, the specific induction of such responses to protection from and elimination of infections, but also cancer and immune-mediated diseases becomes more feasible. Taking advantage of our morbillivirus expertise, we will biotechnologically design morbillivirus-based prototypical vectored vaccines and oncolytic viruses and evaluate their potential to elicit protective immune responses and selectively destroy cancer cells towards clinical applications. In addition, we will apply our animal models to the efficacy assessment of innovative therapeutic strategies and promising vaccine candidates.

4.5. Division of Allergology – Research Towards Improved Diagnosis & Therapy of Allergic Diseases[14]

Head: Prof. Dr. Stefan Vieths

The division is responsible for the regulation of allergen products for both in vivo diagnosis of allergic diseases and allergen immunotherapy (AIT). AIT aims at inducing long-term tolerance by administration of a therapeutic allergen product. Its mechanism of action is not fully understood, but immune modulation including the induction of allergen specific regulatory T cells and immunoglobulin G (IgG) antibodies counteracting disease causing IgE seem to play an important role. Our research focuses on unmet needs in the development of high quality allergen products by regulatory as well as basic research projects.

Clinical trials of novel AIT products generally depend on subjective outcome measures (endpoints) such as symptom scores or quality of life data recorded during field exposure to environmental allergens. These trials often cover two to three years of treatment reflecting the recommendations for clinical practice. As a result, they have a high trial failure risk. We strive to contribute to the evolution of validated endpoints by systematically analysing relevant data in previous regulatory dossiers. Moreover, we support the science-based definition of unmet needs for regulatory acceptance of novel challenge procedures leading to increased trial comparability and reduced variability.

[14] http://www.pei.de/allergology-research

Allergen extracts from biological sources contain a mixture of different proteins including variable amounts of allergenic proteins and isoforms. This leads to particular problems of product quality, safety and efficacy. Hence, allergen product characterization and standardization depends on an accurate determination of potency reflecting the content of clinically relevant allergenic proteins. Current regulatory documents rely on immunoassays with pooled patients' IgE for potency determination in comparison to manufacturer-specific in-house reference preparations of allergen extracts. To support the implementation of more appropriate test systems, we develop immunoassays on the basis of allergen-specific monoclonal antibodies and recombinant reference standard(s) representing defined amounts of relevant allergenic proteins. For expanded innovation of quality control, we work on highly sensitive multi-allergen mass spectrometry assays enabling simultaneous detection and quantification of multiple isoforms of major and minor allergenic proteins in allergen extracts.

In contrast to allergen extracts, recombinant allergenic proteins and derivatives promise the development of novel AIT products predominantly inducing tolerance to clinically relevant allergens. One strategy is the development of hypoallergenic products and synthetic peptides derived from allergenic proteins. These are not recognized by patients' IgE and should hence avoid allergic side effects during AIT. In this regard, we develop and apply advanced methods for systematic identification and characterization of sequential and conformational IgG and IgE binding epitopes of allergens before, during and after AIT. We use the information gained for a targeted design of novel hypo-allergens. In food allergy, we also try to correlate recognition of allergen epitopes by individual patient's IgE to clinical features of disease (e.g. severity or persistence). In summary, these studies support the development of novel approaches for allergy diagnosis and AIT, and will form the scientific basis for their regulatory assessment.

4.6. Division of Medical Biotechnology – Research to Improve the Safety & Efficacy of Advanced Therapy Medicinal Products[15]

Head: Dr. Zoltán Ivics

The division is in charge of clinical trial and marketing authorizations of advanced therapy medicinal products (ATMPs) including gene therapeutics and cell-based products. A special challenge to developers and regulators are severe side effects of

[15] http://www.pei.de/med-biotech-research

ATMPs that often hamper their successful clinical implementation. For example, integration of therapeutic transgene vectors used for gene therapy into a cellular tumor suppressor gene or a proto-oncogene can induce cancer. We implemented a research focus addressing the basic biological and biotechnological principles underlying ATMP development with the goal to aid their translation into the clinic.

To evaluate the risks associated with ATMPs, we develop test systems enabling systematic assessment of the deregulation of patient's gene functions by gene therapeutics, determination of the (epi)genetic stability of cell-based products, and analysis of the cellular signaling pathways influencing the safety of regenerative medicines. Xenotransplantation of animal cells, tissues or organs into humans promises to overcome the critical shortage of allogeneic donors. To contribute to risk assessment regarding cross-species disease transmission, we focus on the biology of endogenous retroviruses in animals. Within the scope of our association to the German Consortium for Cancer Research, we strive to identify and solve regulatory challenges associated with personalized therapies and ATMPs for the treatment of cancer.

Interactions between genomic parasites such as integrating viruses and transposable elements with host cells can have significant impact on the quality and safety of gene therapeutics and cell-based products. For example, when used as vectors in cell and gene therapy, viral- and transposon-encoded factors can have negative consequences on cellular homeostasis, whereas host factors affect the efficacy of gene transfer. To improve the safety and efficacy of ATMPs, we investigate interactions of vector components with cellular factors, for instance by studying chromatin features that determine the patterns of genomic integration of exogenously supplied transgene vectors. In differentiated somatic cells, endogenous transposable elements are transcriptionally silenced. However, their expression is activated in pluripotent cells, which can result in chromosomal integration events. Since these potentially mutagenic events might compromise the biosafety of therapeutic stem cells or their differentiated derivatives, we study (epi)genetic pathways and host-encoded factors affecting the mobilization of endogenous transposable elements.

Random genomic integration of therapeutic gene vectors can lead to serious side effects in gene therapy. To contribute remedies, we develop experimental therapeutic approaches for targeted gene transfer on the basis of viral and transposon-based vector systems. Moreover, we investigate the potential of designer nucleases such as CRISPR/Cas enabling site-specific genome engineering and repair of defective genes. We evaluate these approaches using therapeutically relevant cell types - such as

human T cells - and adequate animal disease models, thereby promoting and accelerating successful clinical translation of gene therapy research.

4.7. Division of Haematology/Transfusion Medicine – Research for Safe Therapies with Blood Coagulation Factors & Blood Cells[16]

Head: Prof. Dr. Rainer Seitz

The division is responsible for the regulation of medicinal products derived from human blood including proteins for replacement therapy, cellular components for transfusion, and haematopoietic stem cells. Our research agenda mainly focuses on scientific issues regarding haemostasis, the arrest of bleeding from an injured blood vessel. Bleeding disorders affecting the blood clotting process are predominantly caused by genetic defects of circulating proteins termed clotting factors (CF). CF replacement therapy is based on the infusion of the required factor as an inactive proenzyme. Upon bleeding it becomes activated to trigger the subsequent proteolytic cascade of downstream CFs, which finally results in blood coagulation and haemostasis.

Both plasma-derived and recombinant blood proteins are per se labile. This problem is aggravated by pathogen inactivation or removal procedures. Moreover, all blood products may contain process- or product-related impurities. Hence, important objectives in testing of blood products are to assure the structural and functional integrity of active substances, and to prove the absence of undesired impurities or modifications. In this regard, we e.g. strive to improve the activity testing of therapeutic von Willebrand factor (vWF) concentrates by developing an in vitro flow-chamber model. This mimics physiological blood flow conditions and will enable functional analysis of vWF-mediated wound adhesion of platelets. We also develop advanced assays for the identification of contaminating proteolytic activities in blood products causing unintended clotting or adverse activation of blood cells. This is accompanied by in-depth root-cause analyses of the underlying mechanisms of production-related CF activation.

A major obstacle in the treatment of haemophilia is the therapy-associated development of alloantibodies (“inhibitors”) by the patient. These inhibitors jeopardize the efficacy of haemophilia treatment by neutralizing the activity of therapeutic CFs. In this regard, we are part of a consortium establishing a Europe-wide haemophilia database as an instrument for the scientific risk evaluation of inhibitor development on

the basis of experimental results and prospective patient cohorts. Building on this, we investigate the mechanisms of and the risk factors for the development of inhibitors in cooperation with the Junior Research Group ”Novel Vaccination Strategies & Early Immune Responses". Our ultimate goals are to contribute to solutions enabling the prediction of the individual patient risk for inhibitor development before the start of therapy, estimation of the respective immunogenic potential of novel (recombinant) therapeutic CFs, and finally evolution of safer and more efficient treatment options.

Clinical and experimental data indicate that the blood coagulation system interacts with tumour cells. To better understand how the tumour benefits from these interactions (such as protection from host defence, generation of a neo-angiogenesis supporting matrix or formation of tumour connective tissue), we specifically study the influence of tissue factor-induced coagulation and cell signalling in a human lung cancer cell model.

4.8. Division of Safety of Medicinal Products & Medical Devices – Research to Identify & Minimize Risks Associated with Biomedicines

Head: Dr. Brigitte Keller-Stanislawski

The division is responsible for the collection, detection, assessment, understanding, and prevention of adverse events associated with vaccines and biomedicines for human and veterinary use (pharmacovigilance). This includes the identification of previously unrecognized adverse events or changes in the patterns of adverse events, and an evaluation of the risk/benefit ratio in order to determine sufficient and proportionate risk minimization measures. In line with this, our research focus aims at rapid detection and analysis of risks, appropriate evaluation of the risk/benefit ratio, identification of the root cause of serious adverse events and proportionate risk minimization.

A basic prerequisite of our pharmacovigilance activities are active and passive surveillance approaches that enable rapid detection and analysis of serious adverse events associated with vaccines and biomedicines in medical use. In this regard, we are developing an electronic surveillance tool which facilitates notification of adverse events by physicians and consumers and guarantees protection of privacy. In a cooperation project with the Helmholtz centre for infection research, we are currently aiming to establish novel tools for easy notification of vaccine-related adverse events.

[16] http://www.pei.de/haematology-research

Risk estimates are based on well-designed pharmacoepidemiological studies to examine the association between the exposure to a biomedicine and the occurrence of adverse events, and to quantify associated risk. To this end, we use a variety of epidemiological study designs for pro- or retrospective analyses of suspected adverse events. Cohort studies to estimate the relative risk compare the risk of adverse events in a group treated with a biomedicine with a homologous control group not treated with this biomedicine. Case-control studies are a useful tool for estimating the frequency of disease. We conduct such studies comparing a group of subjects developing adverse events with a group of non-affected subjects with the aim to examine if the frequency of biomedicine application is associated. Self-controlled case series studies are suited to evaluate whether a vaccine is linked to an increased risk of adverse events if only cases can be investigated (e.g., the occurrence of Gullain-Barré-Syndrom in connection with pandemic H1N1-vaccination). Capture-recapture studies are useful to detect a difference in the incidence of a rare disease before and after biomedicines application (e.g., the incidence of narcolepsy before and after pandemic H1N1-vaccination).

A further important question in pharmacovigilance investigations of biomedicines is to what extent the active substance itself or a pharmaceutical excipient is capable of evoking adverse events. In this regard, we lead pharmacokinetic/toxicokinetic investigations to evaluate specific risks of biomedicine components. One current example is the development of a physiology-based toxicokinetic model to simulate human exposure to aluminium from aluminium-adjuvanted vaccines and allergens. This model shall support, among others, the evaluation of the toxicological risks regarding the specific vaccination scheme for infants.

4.9. President Research Group – Molecular Biotechnology & Gene Therapy[17]

Head: Prof. Dr. Christian Buchholz

Viral gene transfer vectors and oncolytic viruses hold great promise as novel therapeutic tools for a broad variety of devastating diseases. The first essential step for their therapeutic activity is cell entry, which involves attachment of vector or virus particles to cell surface receptors initiating penetration of the cellular membrane and uptake into the intracellular compartment. Particles targeted to therapeutically-relevant

[17] http://www.pei.de/research-groups-president

cells at the level of cell surface receptor attachment promise improved safety and efficacy.

As a partner site of the LOEWE Center for Cell and Gene Therapy Frankfurt and the German Consortium for Cancer Research, we engineer viral vectors and oncolytic viruses carrying specific targeting domains to bind to cell surface receptor proteins of choice. Hence, such receptor-targeted viral vectors (RT-VVs) and oncolytic viruses (RT-OVs) specifically attach to target cells by discriminating between target-receptor positive and negative cells with an antibody-like selectivity. In the future, we strive to better understand the correlation between receptor choice and cell entry routes taken by viruses after receptor attachment.

Based on this, viral vectors targeted to therapy-relevant cell types, such as T lymphocytes, hematopoietic stem cells, and subtypes of endothelial cells or neurons will be generated and their production improved by developing experimental technologies for their purification. We will assess the therapeutic potential of prototypical RT-VVs in neurodegenerative diseases and immunotherapy, the latter with a special emphasis on the delivery of chimeric antigen receptor (CARs) into distinct subsets of T lymphocytes. Prototypical RT-OVs specifically targeted to tumour and tumour stem cells will be evaluated using non-clinical cancer models.

4.10. Vice President Research Group – Molecular Allergology[18]

Head: Dr. Stephan Scheurer

Allergen immunotherapy (AIT) with crude allergen extracts to induce immune tolerance against the allergen is frequently not very effective. Novel treatment strategies using recombinant allergens, adjuvants and/or new administration routes for the development of modular allergen therapeutics (MAT) promise significantly improved efficacy. Our group focuses on the exploration of such novel, more effective MATs and the elucidation of the underlying modes of action.

Innovative MATs combine adjuvant effects with the cell targeting properties of a viral or protein transfer vector. This enables specific targeting of the allergen to immune cells as well as intended modulation of the immune response. Pathogen-associated molecular patterns (PAMPs) are conserved bacterial and viral structures that bind to pathogen recognition receptors on immune cells. PAMPs thereby specifically trigger

[18] http://www.pei.de/ research-groups-vice-president

pro- or anti-inflammatory host immune responses. We exploit PAMPs as adjuvants for targeted immune modulation to allergens by the development of experimental MATs. Our aim is to design novel strategies for the promotion of immune deviation via the induction of allergen-specific IgGs to counteract disease-causing IgEs or the promotion of immune regulation via immunosuppressive regulatory T cells.

MATs should improve therapeutic efficacy, and preferably not induce adverse effects. To this end, we investigate the molecular mechanisms of the immunomodulatory effects of experimental MATs *in vivo* and *in vitro*. These studies will help to disclose their risk/safety profile, which is important information for state-of-the-art regulatory assessment of these innovative biomedicines.

4.11. LOEWE Research Group – Targeted Gene Modification in Stem Cells[19]

Head: Prof. Dr. Dr. Ute Modlich

Stem cells are long-lived, possess self-renewal potential and can be differentiated into all cell types. Stable genetic modification of stem cells with integrating vector systems are maintained in their progenies, and the expression can be controlled to occur in terminal effector cells. This is why stem cells are of particular interest for the development of cell-based biomedicinal products. Since gene vector insertion can lead to undesired cell transformation, we develop preclinical assays for the assessment of the genotoxicity of integrating vectors.

Hematopoietic stem cells (HSCs) give rise to all blood cells, making them an attractive target for cell-based gene therapies for inherited hematological disorders. However, preservation of their stem cell function is especially challenging during ex vivo cultivation and modification. In vivo, the cytokine thrombopoietin (THPO) is essential for HSC maintenance. To improve ex vivo HSC expansion and foster in vivo engraftment of modified HSCs, we therefore investigate THPO-induced genes and pathways as potential therapeutic targets.

The differentiation of blood cells from embryonic stem cells (ESCs) or induced pluripotent stem cells (iPSCs) holds great promise for the production of donor independent blood products. In this regard, we specifically investigate the ex vivo differentiation of platelets from ESCs/iPSCs. Moreover, by genetic modification of

[19] http://www.pei.de/loewe-research

ESCs/iPSCs we aim to generate modified platelets carrying therapeutic proteins to be released locally after platelet activation.

4.12. Junior Research Group – Novel Vaccination Strategies & Early Immune Responses[20]

Head: PD Dr Zoe Waibler

Adverse effects of biomedicines targeting the immune system are an important regulatory issue of the PEI. Based on our expertise in immunology we strive to clarify the underlying immune mechanisms. One example is our ongoing root cause investigation of the severe inflammatory reactions induced by the immunomodulatory monoclonal antibody TGN1412 during the first-in-man study. In a joint project with the Division of Haematology/Transfusion Medicine, we investigate the molecular mechanisms of undesired immune responses upon blood coagulation factor VIII application hampering effective treatment of haemophilia A.

Innovations in the field of vaccines require novel and safe adjuvants to modulate and activate innate immunity in a targeted manner. Detailed insight in early immune responses is therefore a prerequisite for the development of novel adjuvant strategies. Type I interferons are a particularly promising target, since these cytokines are crucial innate immune factors inducing and shaping subsequent adaptive immune responses. For that reason, we aim at elucidating the molecular mechanisms involved in induction, regulation, and inhibition of type I interferons using murine infection models such as mice lacking the type I interferon receptor.

Based on our knowledge on type I interferon biology we also work on experimental vaccination strategies fostering the development of recombinant modular vaccines for infectious diseases, allergies, and cancer. These studies are performed in close cooperation with various PEI research groups possessing specific expertise in the fields of allergen immunotherapy and (tumour) vaccines.

[20] http://www.pei.de/new-vaccination-strategies-research

4.13. Junior Research Group – Cellular Aspects of Host-Pathogen Interactions[21]

Head: Dr. Renate König

Viruses evolve rapidly to evade recognition by the host. This is why hosts must constantly adapt their mechanisms of viral immunity to a moving target. This co-evolutionary arms race has led to a highly refined interaction between viruses – which co-opt cellular host dependency factors (HDFs) essential for their replication – and host cells – which develop pattern recognition factors (PRFs) and antiviral restriction factors (ARFs) to detect and combat virus infection.

Viral evasion of immune recognition frequently hampers the potency of vaccines. This is especially true for highly variable RNA viruses. We strive to discover intracellular host cell factors influencing viral replication either positively (HDFs) or negatively (PRFs and ARFs) and wish to elucidate their function in detail. Our ultimate goal is the design of improved vaccines with innovative adjuvants to trigger the initial immune response and promote and shape adaptive immunity.

To this end, we employ a systems-based high-throughput screening approach using arrayed libraries to either up-regulate or silence host cell gene expression prior to viral infection. Up-regulation is achieved by using focused libraries of copy-DNA (cDNA), such as those for interferon-stimulated host genes. Silencing is realized by means of genome-wide RNA interference (RNAi) libraries to inhibit the translation of host cell messenger RNA into protein. We manipulate host cell gene expression gene by gene and examine whether it has an effect on viral replication or the virus specific innate response. High content screening technology enables us to elucidate the actual stage of the virus replication cycle and gives rise to a deeper understanding of the underlying mechanisms.

5. Outlook

This outlook gives a short overview of mid-term and long-term challenges requesting additional research efforts by our scientists to meet important regulatory or societal needs.

[21] http://www.pei.de/host-pathogen-interactions-research

In the light of our participation in the German Centre for Infectious Disease Research (DZIF), we face the development of entirely new modular vaccine platforms for the prevention of infectious diseases. These products cannot be approved under the regular regimen of assessment procedures for common vaccines, but require innovations in the evaluation criteria and processes for the assessment of clinical trial and marketing authorisation applications. A particular challenge consists in establishing the scientific basis for quality and clinical acceptance criteria. Here, conjoint interdisciplinary research efforts involving our regulators and research scientists are required for developing adequate solutions.

Another midterm challenge is the need for innovation of experimental test procedures as well as improvements in the assessment of quality, efficacy, and safety of vaccines. In this regard, we will establish an interdisciplinary cooperation of regulators and research scientists with expertise in the fields of vaccines assessment, vaccines development, batch release testing, and non-clinical models. This initiative is aimed at discovering and validating novel correlates of protection, and to foster the development of relevant and sufficiently human-like animal models for an improved preclinical assessment.

A third mid-term need results from the experiences made during the 2014 Ebola outbreak. In this context, we learned that one of the major challenges was to have an emergency treatment including candidate vaccines at hand. International and national coordination of indispensable regulatory consultation processes turned out to be highly complex, and engagement of pharmaceutical companies was limited. Protecting the human and animal population from new and re-emerging infectious diseases remains hence an urgent societal need. In this regard, we strive to use our regulatory expertise for the development of harmonized regulatory procedures enabling, for instance, fast track approval of clinical trials for emergency treatments. As part of our long-term strategy we also aim to use our research expertise for anticipatory development of such emergency treatments.

Vaccines and biomedicines regulated by the PEI are the most promising candidates for the prevention and treatment of neglected infectious diseases, rare genetic disorders and tumours, chronic inflammatory diseases, and allergic diseases caused by uncommon antigens. Pharmaceutical companies are per se not in the position to promote treatment solutions for such neglected or uncommon diseases. As a federal regulatory authority and research institute, the PEI is committed to contributing to progress in this field by regulatory and research efforts, thus developing the PEI into

one of the leading European institutions in the prevention and treatment of neglected diseases.

Prof. Dr. K. Cichutek
President of the
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