Newly discovered rare disease helps understand the complexity of bowel homeostasis

A single-gene mutation may cause a severe bowel disease in children. This could be shown in a recent study by an international research team[1] led by Kaan Boztug at CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences and the Ludwig Boltzmann Institute for Rare and Undiagnosed Diseases (LBI-RUD) together with Michael Lenardo, National Institute of Health (NIH), Bethesda. The newly discovered alterations lead to total loss of a protein called CD55, causing life-threatening bowel inflammation, chronic diarrhea and vascular thrombosis. The researchers unraveled the underlying molecular mechanisms and discovered previously unknown regulatory pathways relevant to bowel homeostasis. As a consequence of their findings, the researchers could identify a clinically approved drug which may specifically interfere with aberrant signaling involved in this previously unknown disease. The manuscript has now been published in the renowned New England Journal of Medicine.

(Vienna, June 29, 2017) Among the plethora of genetic alterations, those causing a complete block of protein production belong to the most severe mutations. When a stopping signal, a so-called *stop codon* is inserted into a protein's genetic blueprint, the resulting protein products are dysfunctional. Considered useless, they are usually guided to a cell's degradation machinery – a process that potentially causes disastrous consequences for an organism.

In a young female patient suffering from early-onset inflammatory bowel disease and protein-losing enteropathy, such a severe type of mutation was identified by the team of Kaan Boztug, director of the newly founded LBI-RUD and affiliated principal investigator at CeMM and the Medical University of Vienna. The resulting study was recently published in the New England Journal of Medicine (www.nejm.org).

The little girl who had undergone numerous hospitalizations and therapeutic attempts suffered from various symptoms from early childhood, ranging from severe diarrhea to recurrent infections, chronic malnutrition and concomitant failure to thrive. With a genetic investigation, a so-called exome sequencing, which identifies and reads all protein-encoding genes, Kaan Boztug´s group could shed light on the clinical mystery: Rico Ardy, a PhD candidate in Boztug's team, could identify a stop codon within the gene sequence of the protein CD55, which is a key

[1] with contributions by the National Institutes of Health, USA, and the University of Istanbul, Turkey

regulator of the innate immune system. Yet a definite proof of whether this single mutation was causal for the complex disease manifestation of this unique patient needed even more detailed molecular investigations.

At this stage, Kaan Boztug's involvement in international networks of researchers working on rare diseases proved beneficial: Michael Lenardo, long-standing collaborator at the US-based National Institutes of Health (NIH), had independently identified two other families with index patients exhibiting similar clinical symptoms who also carried inherited mutations in the *CD55* gene. Subsequently, the research team which was jointly led by Kaan Boztug and Michael Lenardo was able to identify a total of 11 patients, and addressed the detailed molecular mechanisms: Absence of CD55 protein dampened a proper control of the complement system, an important component of innate immune defense, inducing proinflammatory cytokine signaling. Concomitantly, the researchers could show that loss of CD55 led to impaired production of IL-10, a key anti-inflammatory signaling molecule in the gut.

The combination of these effects lead to a severe clinical disease which may be life-threating: The over-functioning complement system collapses in the absence of regulatory IL-10 molecules. The observed bowel inflammation is an immediate consequence, enforced by the constant immune cell stimulation through the gut flora, where fine-tuned regulatory mechanisms are of particular relevance. Inflamed intestinal lymphatic vessels lead to loss of protein through blood plasma, a major disease manifestion of CD55 deficiency, accompanied by a risk of thrombosis.

After deciphering the molecular consequences of this particular genetic defect, the investigators aimed at identifying a potential targeted therapy for this condition, and were successful: A clinically approved drug that dampens complement activity could also be applied in case of absent CD55 protein.

"This disease is an ideal example for the importance of rare disease research," says Kaan Boztug. "By investigating the molecular mechanisms underlying the disease, we are then able to strategically test for re-purposing approved medications for a personalized therapy. Furthermore, through investigating single genetic defects, we are able to unravel novel aspects of the biological function of the affected gene."

The role of CD55 in maintaining bowel homeostasis was hitherto largely unexplored. Similarly, the molecular mechanisms underlying protein loss in the bowel – a process that plays a role in a wide array of diseases – were widely unknown. Here, the present study provided important insights that widen our understanding of the complex regulatory mechanisms of bowel homeostasis, which could eventually be relevant for other types of rare and common diseases.

The study "CD55 Deficiency, Early-Onset Protein-Losing Enteropathy and Thrombosis" was published on June 28, 2017 in the *New England Journal of Medicine*. http://www.nejm.org/doi/full/10.1056/NEJMoa1615887#t=article

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Kaan Boztug studied human medicine in Düsseldorf, Freiburg and London, after which he graduated with Ian Campbell at the Scripps Research Institute (La Jolla, US). He completed his clinical training and postgraduate research with Christoph Klein at Hannover Medical School prior to his first appointment as an independent group leader in 2011 at the CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences. Since 2011, he holds a dual appointment as Associate Professor at the Department of Paediatrics and Adolescent Medicine at the Medical University of Vienna. Since 2016, Kaan Boztug is director of the Ludwig Boltzmann Institute for Rare and Undiagnosed Diseases. He is furthermore director of the CeRUD Vienna Center for Rare and Undiagnosed Diseases as well as the Jeffrey Modell Diagnostic and Research Center Vienna at the Medical University of Vienna and the St. Anna Childrens' Hospital.

The **CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences** is an international, independent and interdisciplinary research institute for molecular medicine, directed by Giulio Superti-Furga. CeMM's mission is driven by unmet medical needs and integrates basic research and clinical expertise to develop innovative diagnostic and therapeutic strategies for precision medicine. Its research focuses on cancer, inflammation, metabolic and immune as well as rare diseases. The research building is integrated into the campus of the Medical University of Vienna and the Vienna General Hospital.
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The **Ludwig Boltzmann Institute for Rare and Undiagnosed Diseases (LBI-RUD)** was launched by the Ludwig Boltzmann Gesellschaft in April 2016 together with its partner institutions including the CeMM Research Center for Molecular Medicine of the Austrian Academy of Sciences, the Medical University of Vienna and the Children's Cancer Research Institute and the St Anna Children's Hospital Vienna. Its research remit is the thorough analysis of rare diseases of the hematopoietic system, the immune system and the nervous system – as

such not only dedicated to provide research for the development of personalized therapeutics for affected patients, but with similar efforts dedicated to unravel novel insights into human biology. Benefitting from full access to the infrastructure of its partner institutions, LBI-RUD has established a coordinated research programme, integrating and considering scientific, sociologic, ethical and economical aspects of rare diseases.
www.rare-diseases.at

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