Dr Daniel Globisch

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Analysing gut microbiome metabolites to detect disease

Research Objectives

The development of new chemical biology-based methodologies to improve the analysis of small molecule metabolites in biological samples.

Detail

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Bio
Associate Professor Daniel Globisch has his laboratory at Uppsala University, Sweden, with a research line in chemical biology and metabolomics. He has a PhD from the University of Munich, Germany, and did his postdoctoral training at The Scripps Research Institute, La Jolla, USA, 2011–15.

Funding
- Vetenskapsrådet
- Cancerfonden
- SciLifeLab

Collaborators
- Professor Matthias Löhr

References

- Lin, W, Conway, LP, Vujasinovic, M, Löhr, JM, Globisch, D, (2021) Chemoselective and Highly Sensitive Quantification of Gut Microbiome and Human Metabolites. *Angewandte Chemie (International ed. in English)*, 60(43), 23232–40.
- Conway, LP, Garg, N, Lin, W, et al, (2019) Chemoselective probe for detailed analysis of ketones and aldehydes produced by gut microbiota in human samples. *Chemical communications (Cambridge, England)*, 55(62), 9080–83.
- Ballet, C, Correia, M, Conway, LP, et al, (2018) New enzymatic and mass spectrometric methodology for the selective investigation of gut microbiota-derived metabolites. *Chemical science*, 9(29), 6233–39.
- Garg, N, Conway, LP, Ballet, C, et al, (2018) Chemoselective Probe Containing a Unique Bioorthogonal Cleavage Site for Investigation of Gut Microbiota Metabolism. *Angewandte Chemie (International ed. in English)*, 57(42), 13805–9.

Personal Response

How soon are you planning to start a study with a larger cohort of patients?

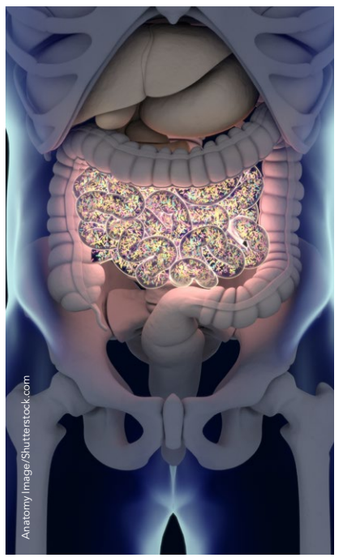
"The samples are currently being collected by our medical collaborators and we will perform this analysis after we have received these. We are aiming for long-term monitoring of variations of these compounds over time to identify specific metabolic signatures that can predict disease development and progression

Are there any diseases you have in mind that can be diagnosed through your method?

The method has been developed for broad application to complement any metabolomics-based biomarker discovery projects. I am hopeful that our method will find broad application in metabolomics research to obtain a more detailed overview of metabolic changes in the context of any disease. Furthermore, projects that aim to identify differences in microbiome metabolism in two different cohorts could also benefit from our method."

Analysing gut microbiome metabolites to detect disease

Alterations in the gut microbiome can lead to the production of highly reactive compounds that can contribute to disease development. Dr Daniel Globisch and his team from the Department of Chemistry at Uppsala University, Sweden, have developed a chemoselective probe immobilised to magnetic beads for the analysis of microbiome metabolites in complex human samples. This novel mass spectrometric method is a new tool towards personalised medicine and diagnostics, making it possible to monitor patients and how their disease is progressing through time.



An unquantifiable number of micro-organisms live on and within the human body, particularly the gastrointestinal tract – known collectively as the microbiome. These microbes are referred to as 'commensals', as they 'eat from the same plate', exchanging metabolites with their host. The genetic pool of commensal micro-organisms contains about 400 times more genetic information than the human genome itself, and many of these genes encode for the metabolism of compounds whose physiological role is yet to be discovered.

While commensals benefit their host by offering protection against pathogens, abnormal alterations in the microbiome and its metabolism have been linked to disease development. There is increasing evidence that a poorly regulated gut microflora makes a significant contribution to a variety of diseases, including cancer, diabetes, obesity, cardiovascular diseases, and inflammatory bowel disease. Our knowledge of the overall metabolic interactions of commensal microbial communities with the human host is still limited, preventing us from fully evaluating the toxic or beneficial properties of these metabolites.

NOVEL ANALYSIS OF BIOLOGICAL SAMPLES

Metabolomics aims to investigate all of the metabolic products present in a biological sample. Samples can either be analysed by nuclear magnetic resonance or by mass spectrometry. Human urine and plasma samples are increasingly being used as sources of biomarkers in the form of 'liquid biopsies', which have the advantage of being readily accessible without the need for invasive and complex procedures. While plasma samples provide real-time information of ongoing metabolic pathways, urine samples contain metabolic end-products that are cleared and excreted by the kidneys.

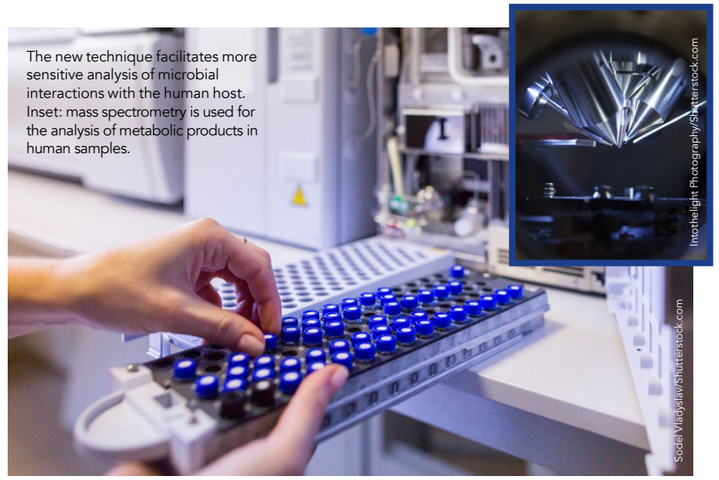
Dr Daniel Globisch and his team from the Department of Chemistry at Uppsala University, Sweden, have developed newly sensitive analytical methods to monitor metabolites produced by the gut microbiome, in order to identify biomarkers that might indicate the onset of a disease and its progression over time.

ENZYMATIC PRE-TREATMENT FOR TARGETED METABOLITE ANALYSIS

In 2018 the team published a study describing the targeted metabolite analysis of biological fluids, using sulfatase pre-treatment followed by analysis using mass spectrometry. The study reported the successful identification of 206 sulfated metabolites in human urine and faecal samples, and on the discovery of a large number of previously unreported metabolites.

Metabolic enzymes, such as cytochrome P450 oxidases, introduce charged groups into xenobiotics, including microbiota-derived metabolites, to facilitate their excretion from the human body. This process, however, can also lead to bioactivation and the generation of toxic compounds. The method reported by Globisch's team enabled the detection of the sulfate ester and the identification of its structure. The sample was then split in two parts: the first was treated with sulfatase, while the second had inactivated enzyme, acting as a baseline to exclude any signals not resulting from enzymatic treatment.

Once the baseline was subtracted, the authors concluded the approach could be used to discover metabolomics-driven biomarkers to enable detection of diseases affected by changes in the composition of the microbiome.



The new technique facilitates more sensitive analysis of microbial interactions with the human host. Inset: mass spectrometry is used for the analysis of metabolic products in human samples.

ANALYSING MICROBIAL INTERACTIONS WITH QUANT-SCHEMA

In their new research, Globisch and his team have developed a technique that enables the detection of more than 200 metabolites in human faecal, urine, and plasma samples, facilitating more sensitive analysis of microbial interactions with the human host and the role they play in disease development.

The team has developed a sensing probe immobilised to magnetic beads for the mass spectrometric analysis of metabolic products in human samples: Quantitative Sensitive CHEmoselective MetAbolomics (quant-SCHEMA). The technique allows the simultaneous analysis of two sets of samples, one pre-treated with a stable isotope-labelled tag and another with a 'light' tag, which is devoid of isotopes. 'This is a unique approach,' explains Globisch, 'as other methods lack separation from the sample matrix and must overcome matrix interferences.' This comparative analysis has the advantage of reducing mass spectrometric background interference and enhancing the level of detection by up to six orders of magnitude, allowing the researchers to detect metabolites at incredibly low concentrations. The increased sensitivity of the probe is also possible because of the immobilised magnetic beads which efficiently separate captured metabolites from the sample matrix, which is the main cause of ion suppression.

The probe enabled the researchers to validate 60 metabolic products from faecal samples. Some of the products, which were either of microbial origin, food products, or endogenous metabolites, have previously been linked to the development of disease. For example, 5-Keto-D-gluconic acid is a known metabolite from the Gluconobacter genus and is produced from D-gluconate metabolism through catalysis by the enzyme D-gluconate dehydrogenase (GADH). Other examples include pyruvic acid and 4-hydroxyphenylpyruvic acid that are excreted from the common gut bacterium *E. coli*. The volatile compounds acetaldehyde and acetone are common endpoints of many biochemical pathways in the metabolism of the commensal organisms. These compounds have been associated with the development of a broad range of diseases and demonstrate the diagnostic value of the procedure, which can track changes in the concentrations of biologically relevant molecules.

DETECTING DISEASE BIOMARKERS

The team's new study, published in 2021, demonstrates the application of the quantSCHEMA method for analysing carbonyl compounds in human faecal, plasma, and urine samples collected from patients with pancreatic cancer. 'The analysis and quantification of this compound class remains challenging due to its chemical properties', says Globisch. The team's analysis led to the discovery of a series of previously unknown metabolites of this class of reactive compounds, as well as providing some insights into carbonyl regulation in humans. The study revealed distinct pattern in the metabolic products detected, which acted as distinct 'chemical signatures' for the individuals, revealing important information about their health. A group of five metabolites including butanal, acetaldehyde, acetone, valeraldehyde, and diacetone alcohol, followed the same changes in plasma and urine over time. The quantSCHEMA methodology enables this reactive compound class to be measured in urine and plasma samples over time to monitor quantitative alterations. Using this new technique, the researchers were able to identify some important individual differences. At the second timepoint in the study, four metabolites (hydroxyacetone, glycolaldehyde, acetylacetone, and acetoin) were upregulated in urine and plasma samples for all patients. The five microbial metabolites acetaldehyde, acetone, pyruvic acid, 4-hydroxyphenylpyruvic acid, and 5-keto-D-gluconic acid present in faecal samples were also detected in plasma and urine samples.

Abnormal alterations in the microbiome and its metabolism have been linked to disease development.

The specific individual metabolic changes identified in the 2021 study could now be monitored in larger groups of patients to identify the key signatures of disease onset and development. The team anticipates that their quantSCHEMA method will improve understanding of how microbial metabolic interactions with the human host can lead to disease progression. This will be an important step towards the development of a detailed 'personalised medicine' profiling.

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