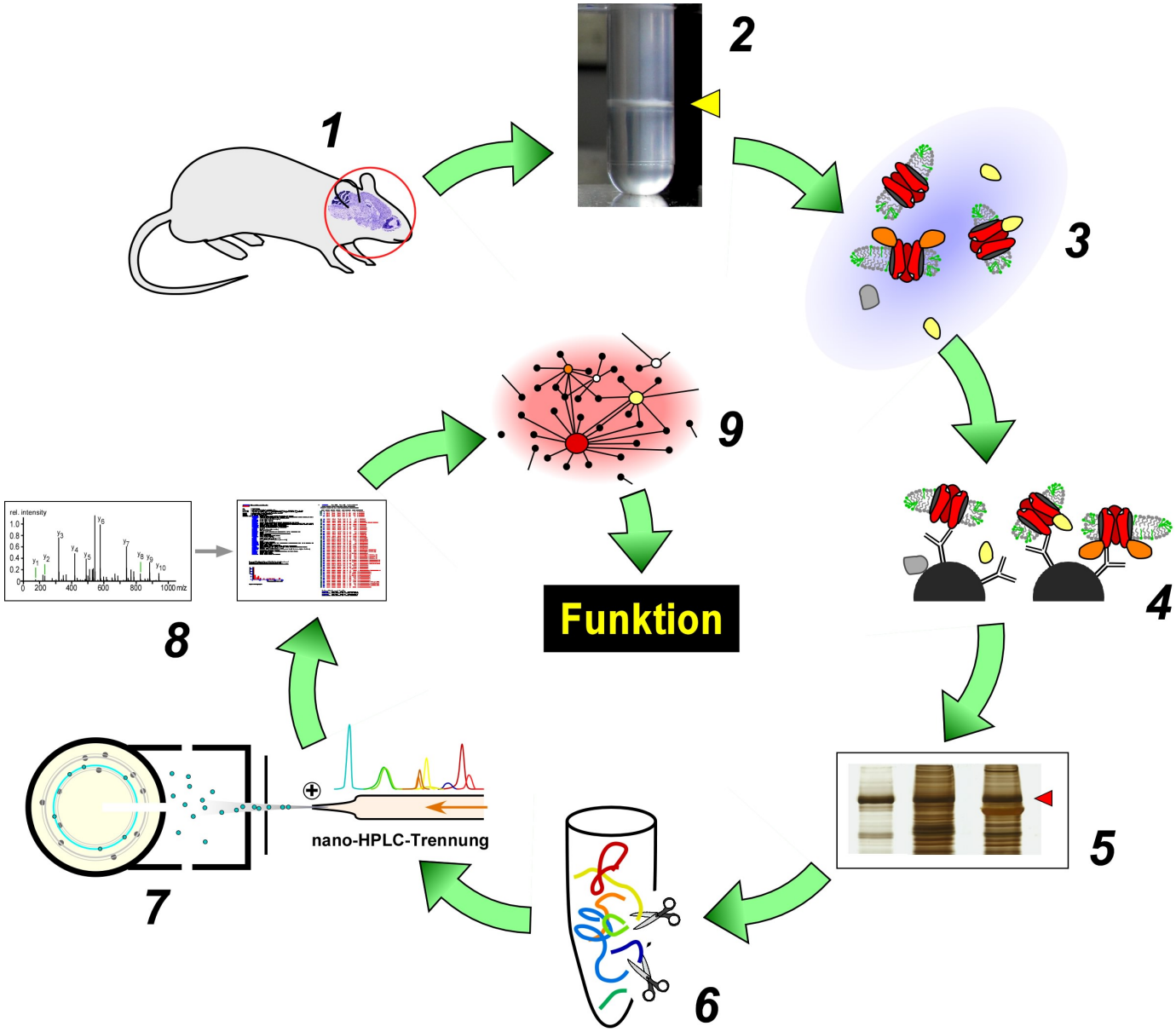
Healthcare industry BW

From protein complex to network

“In order to fully understand an organism, it is necessary to consider it as a whole,” said Dr. Uwe Schulte, biochemist and CEO of Logopharm GmbH in Freiburg. A growing number of scientists hold the same view, and there is a growing inclination to research the big picture. This is reflected in the continuously increasing number of projects looking into systems biology research.

About ten years ago, the majority of scientists still believed that they would be able to decipher the secret of life once they had identified all the genes in a particular organism. However, growing insights into biological processes have shown that this is not the case. “The numerous interactions between the gene products (proteins) form the true basis of biological processes. And these interactions cannot be deduced from genetic information (DNA),” said Schulte. Logopharm GmbH, a specialist in the isolation and characterisation of protein complexes, is taking this paradigm change into account. Whereas the Logopharm team, on behalf of academic institutions, biotech and pharmaceutical companies, previously focused on the interactions between individual membrane proteins and the interactions of membrane proteins with proteins in the cell’s interior, the company now has plans to expand its membrane proteomics platform and decipher the interplay of proteins in large networks.



Proteomics strategy for the identification of protein funtions: 1 tissue preparation; 2 isolation of membranes (density gradient centrifugation); 3 solubilisation of protein complexes with gentle detergents; 4 affinity purification of (complex) target proteins with antibodies; 5 SDS page separation and staining of isolated proteins; 6 tryptic digest of peptide fragments; 7 mass spectrometric analysis of peptides (nanoLC-MS/MS); 8 protein identification through database search; 9 identification of protein-protein interactions and interaction networks.

Identification of the individual partners of a protein complex can be tedious and time-consuming. “Nowadays, we usually only need a few months to identify a target protein’s interaction partners; but the functional characterisation of the protein might take several years of hard work,” said Schulte. At first, scientists have to carefully remove the intact protein complexes from carefully prepared membranes. Specific antibodies are then used to identify the target proteins, which are subsequently degraded into peptides. The peptides are subsequently analysed and quantified with mass spectrometers. Schulte is well acquainted with these processes and knows that a lot of experience and time are required to develop functional assays for new interaction partners. The assays will then provide researchers with information about the interaction of the identified proteins, their function within a particular complex and their interactions with foreign substances.

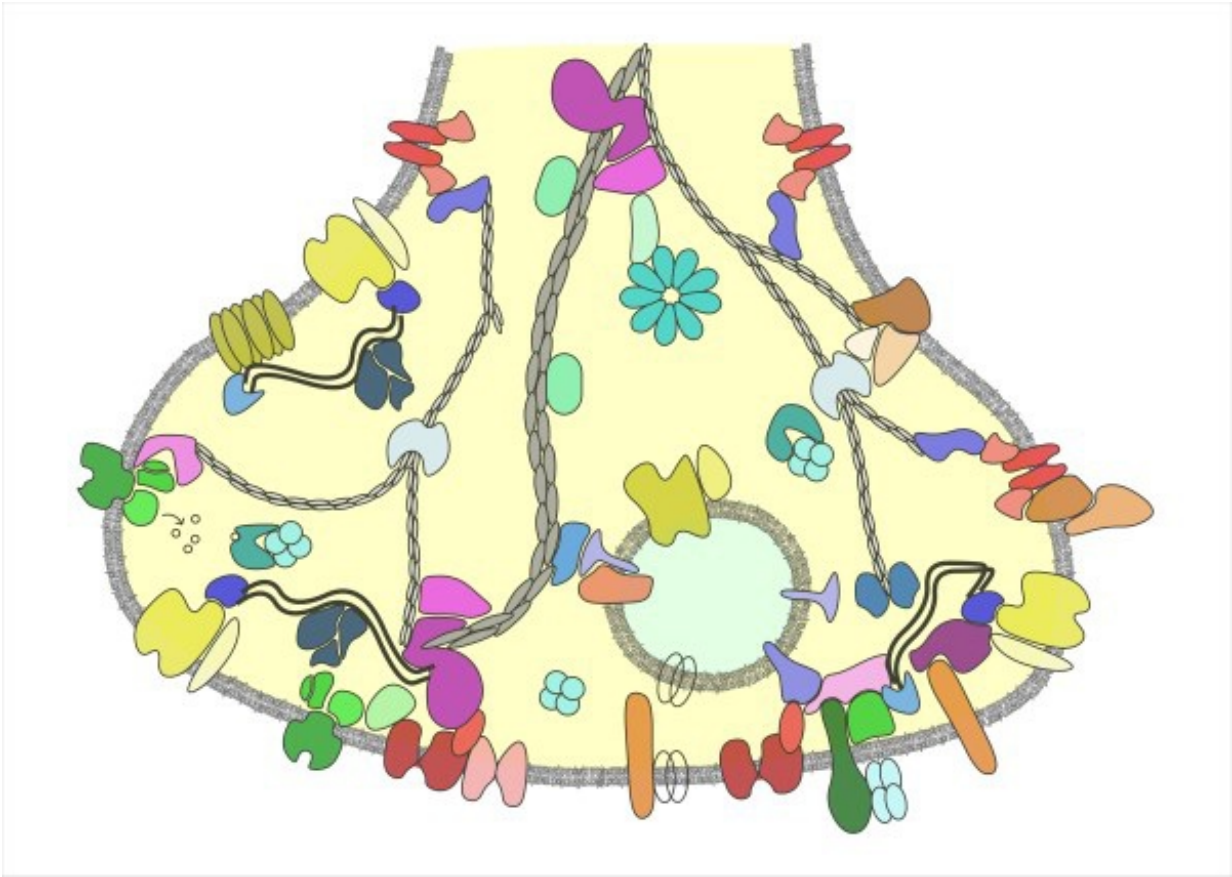
New target structures for drugs

Schulte is no fan of using high-throughput methods to produce huge amounts of data which are difficult to assess. Logopharm does not use strategies that are only able to identify a specific part of the important reaction partners. “One never knows what happens and which function of the protein gets lost,” said the CEO explaining that proteins that influence each other in the complex are the rule and not the exception. “Many important properties of a protein are also determined by the other partners that are part of the complex,” said Schulte. In terms of pharmaceutical research, the conclusion should therefore be that new drugs should not target individual proteins, but protein complexes or even entire networks. “Many substances are known to bind excellently to an individual protein in in vitro tests, but fail almost completely when exposed to a native protein complex,” said Schulte.

The detailed knowledge of protein complexes and networks enables the development of very specific substances that are therefore more selective at the same time as leading to fewer undesired side effects. It is therefore possible to look for substances that do not compete with important biological binding partners of the complex or network. Such knowledge is also of great importance for the production of reliable diagnostic tools or screening systems.

Made-to-measure assays

Schulte’s team has a great deal of experience in the examination of ion channels. “In many cases it would be useful to be able to block certain ion channels,” said Schulte. The original idea was that the pores through which the different ions cross the membrane are ideal targets for new active substances. However, since the pores of the different ion channels have a similar structure, this approach has often failed in practice. “The new approach is to not block the channel pore directly, but to target mechanisms that control the opening and closing of the pore,” said Schulte.



Chemical information is translated into electrical information at synapses – this is a process that involves many helpers: cytoskeleton proteins (black/grey filaments), adaptor proteins (ultramarine blue) and membrane anchors (brown, orange) hold the functional units of the synapse together. These include receptors (green), ion channels (red) and membrane transporters (yellow-green). Under the control of modulators (cobalt blue), "scaffold" proteins (lilac) link the different components together, Enzymes are catalysts in the living cells. They allow the execution of chemical transformations of the metabolism at body temperature.enzyme complexes (bluegreen) support the metabolism and regulate protein functions.

The most important thing in the early stages of the drug discovery process is the establishment of suitable test systems. Assays have to be adapted to specific uses, i.e. not only for an isolated target protein, but also for a target protein complex or, even better, for a complex network. The technology developed by Schulte and his team is able to identify the relevant physiological partners. Over the last few years, Logopharm GmbH has demonstrated the competence behind its membrane proteomics platform in the deciphering of a broad range of protein complexes. In the future, the Logopharm team will use its knowledge for the deciphering of more branched networks.

Junctions to be focused on by the researchers

The researchers will focus on the functional subunits of membrane channels and receptors as well as on the junctions within the signalling pathways involved. Potentially, these subunits and junctions might be the most useful targets for new drugs," said Schulte explaining that it is usually best to look at only one specific receptor in a large network. This can be best explained using tumour cells as an example. In the attempt to stop the uncontrolled growth of tumour cells, it seems plausible, at least initially, to block the binding sites of growth-stimulating receptors. However, this approach often fails because other receptors can also produce an effective growth signal. Sometimes, entire pathways in a network have a parallel redundancy. If, for example, a drug blocks the key protein of the main signalling pathway, then the signal can still be transferred to the target along a byway.

The Logopharm team has recently started to investigate larger networks and has plans to systematically and broadly analyse such networks. The researchers hope to be able to describe and analyse the network partners as comprehensively as possible. Schulte's objective is clear: "We hope to gain a deeper understanding of biological processes."

kb - 2 Oct. 2008

Further information:
Logopharm GmbH
Dr. Uwe Schulte
Schloßstr. 14
79232 March
Tel.: +49 (0)7665/9343-40
Fax: +49 (0)7665/9343-70
E-mail: info@logopharm.com

Press release

17-Oct-2008