

SysBioMed Workshop on Systems Biology for Basal Ganglia Disorders (BGD)

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Participants

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Presentations

First Nicolas LeNovère introduced to the functions and dysfunctions of basal ganglia and the biochemistry and pathology of basal ganglia disorders (BGD). These disorders share the same anatomical substrate, namely a brain structure composed principally of the mesencephalic dopaminergic nuclei, the striatum, and the pre-frontal cortex.

The most important BGDs fall into this category:

- Parkinson's disease (PD)
- Huntington's chorea
- Hemibalism
- Schizophrenia
- Drug addiction
- Depression
- Obsessive-Compulsive Disorder
- Tourette syndrome
- Attention-Deficit/Hyperactivity Disorder (ADHD)

Although both their pathogenesis and their symptoms are very different, the technological bottlenecks faced when developing a systems biology approach are similar.

Concludingly, he briefly looked at the diverse approaches in modelling: electrical modelling (neural-network, cable approximation), chemical kinetics (deterministic, stochastic ...), reaction-diffusion (PDE, finite elements ...), single particle dynamics and others.

Then the participants presented their research focus in concise lectures of 20 minutes each.

Jochen Prehn is developing biological models of neurodegenerative disorders. He presented examples of *in vitro* and *in vivo* models suited to study the degeneration of dopaminergic neurons of the *substantia nigra* which is relevant to PD. *In vitro* models (Primary dopaminergic neurons from mice or rats, cultured as dissociated cells or in organotypic cultures; PC12 cells, neuroblastoma cells or other transformed cell lines; neural stem cells, embryonic stem cells) and single cell imaging deliver good data for systems analyses: quantitative, with good to excellent temporal and spatial resolution. Moreover, significant events can be averaged out in population studies. His *in vivo* studies employ MPP+ / MPTP lesions as well as 6-OH-Dopamin lesions in rodents and other species; alpha-synuclein transgenic animals, and Omi/HtrA2 knockout mice.

Serge Schiffmann presented the current state of the development of *in vivo* models of some neuronal disorders. The utility of any animal model critically depends on the experimental level of analysis, with a multilevel approach more likely to identify novel features. Generating bona fide mouse models for schizophrenia and most human psychiatric disorders is complicated. In comparison to neurodegenerative diseases like Alzheimer's disease or amyotrophic lateral sclerosis, (+PD, HD) where there is a well defined neuropathology, neuropathological markers are not readily apparent in most psychiatric disorders.

Schiffmann presented animal models of drug addiction, KO & transgenic mice with altered responses to psycho-stimulants. Sensitization is achieved by drug self-administration and intracranial self-stimulation. Conditioned place preference (CPP) is regarded as "readout". He also introduced knockouts of mouse orthologues of schizophrenia susceptibility gene candidates which serve as rodent models of schizophrenia. If such models are suited to elucidate the etiology remains to be investigated. Knockout mice with candidate genes affected also serve as animal models of depression. They show physiological changes and patterns of transmitter concentrations and regional brain activity resembling those associated with the disorder in humans.

Guus Smit presented "Novel avenues in high-throughput proteomics and functional analysis of genes". His group performs HT proteomics experiments to study the dynamics of the proteins of the synaptic membrane. The synaptic membrane fraction proteome – the proteins mainly synthesized at the synapse from a local pool of mRNA - consists of roughly 900 proteins. Smit investigates drug addiction and the effects of calcium/calmodulin-dependent protein kinase II (CaMKII) at the proteome level. CaMKII is a molecular switch involved in the storage of long-term neural changes. The activity of the CaMKII holoenzyme (the complete enzyme consisting of both regulatory and catalytic subunits) is controlled by its autophosphorylation state. His group is also working on the identification of genes associated with neuronal regeneration by analysis of the effects of transcription factor mutants. As automation is indispensable in standardization and validation of high throughput phenotyping (HTP) Guus Smits laboratory developed Phenotyper, an automated system for the analysis of cage behaviour suited to detect the aberrant behavior of mouse mutants.

Nicolas Le Novère presented the modelling of neuronal function and molecular and sub-cellular levels. After listing the different processes to model to understand basal ganglia disorder: Electrical properties, chemical interactions (metabolic networks, signalling pathways, neurotransmitter receptor function, gene transcription; protein translation), molecule movements, ionic diffusion, protein transport, compartment remodelling, he presented some of these using examples from his group. The model of chemical interactions was illustrated through the interaction of dopamine and glutamate interactions. Several papers were recently published on the topic, showing that this kind of approach is gaining momentum. The problem of small numbers, combinatorial explosion and spatial localisation were tackled using agent-based modelling, through the modelling of calcium/calmodulin multistate behaviour and the diffusion of receptors in the membrane. Many approaches have been developed that allow us to address every questions asked above, and the bottleneck remains the availability of quantitative data of high quality.

Upinder Bhalla discussed the modelling of whole neurons. Neurons are among the best modeled entities in biology. There are five major aspects to consider:

- Passive electrical conduction, - Synaptic input, - Active properties: voltage-gated channels, - Calcium influx, - Intracellular signaling. Different approaches have been made of which passive compartment models that 'chop' the neuron into little cylinders are well suited to study electrical properties (cable equation). They deliver real-time detailed models (100 compartments, lots of channels, Ca) and 10,000 compartment models with ~100,000 ion channel instances. Electrical models also provide input to chemical models where diffusion is represented as flux of molecules between compartments. Further refinement comes from mass-action kinetics (reaction-diffusion model). There are still a lot of technical problems to solve, i.a. the mismatch of time-scales: electrical changes occur within < msec while chemical steps are in the 1 sec range which would require variable timesteps in modelling. However, one of Bhalla's models was able to demonstrate the neuronal signal transduction as the spreading of an activity pattern along the chain of compartments.

Kevin Gurney, one of the pioneers of the modelling of basal ganglia, presented a 'computational hypothesis': The basal ganglia play a critical role in solving the problem of action selection in animals. Mechanistically, the basal ganglia is a central 'switch' that grants access to motor resources from action requests with high urgency or salience where 'actions' can include overt motoric behaviour and 'thoughts' and 'cognitions'

The selection hypothesis is a unifying one: other functions are not excluded, rather they are reinterpreted in terms of selection. In other words: Disorders of the basal ganglia are disorders of action selection. In this model, the input-output characteristics are those required to select, and switch between actions, the dopaminergic modulation being consistent with PD and some other pathologies. Gurney also presented a robot model of a rat that picks up food and hides in a corner to eat. The control architecture of the robot decomposes into 'behavioural units' (wall-see, corner-see, can-pick-up), rather than functional units (perception, planning, motor control).

Jürgen Gallinat is investigating the role of dopamine and glutamate in the patho-biology of schizophrenia. He employs nuclear magnetic resonance imaging for the detection of glutamate in the brain and finding of correlations of glutamate concentrations and neuropsychological test performance. One found that the cerebral glutamate concentration in schizophrenia is reduced in the anterior cingulate cortex while increased in the hippocampus. Combining these results with the vivo activity of the ventral striatum ('reward system', associated with euphoric feelings) showed that reward-related activation of the v. striatum is reduced in unmedicated schizophrenic patients, also reduced in patients with typical vs. atypical Antipsychotics, and increased in alcoholics in response to alcohol cues.

Jens Pahnke reported on Parkinson syndromes, histological and clinical subtypes, and new approaches in the pathogenesis research of Alzheimer's disease (AD) and PD. One of the most promising therapeutic approaches to PD is deep brain stimulation with micro-electrodes which stimulate the *nucleus subthalamicus* region. It can be considered a systems approach as it affects a key regulatory functional unit of the brain. Pahnke explained that the pathogenesis of AD is likely due to inhibition of protein transport by ABC transporters eventually resulting in aggregation of amyloid proteins and impaired regeneration of brain cells. There is a wide field for the application of bioinformatics methods to investigate the clearance mechanisms at the blood-brain barrier which separates brain cells from blood vessels, especially the kinetics of clearance vs. protein aggregation are subject to modelling.

Jan G Bjaale represented the International Neuroinformatics Coordination Facility (INCF), an OECD initiative. It aims to coordinate and foster international activities in neuroinformatics, to contribute to development and maintenance of database and computational infrastructure and to support mechanisms for neuroscience applications. Main goal is the advancement of neuroinformatics - an interdisciplinary research area, combining research in neuroscience and informatics (including computation) to develop and apply advanced tools and approaches needed for understanding the brain. INCF seeks to enable access to all freely accessible data and analysis resources for human brain research to the international research community. The organisation also develops mechanisms for the seamless flow of information and knowledge between academia, private enterprises and the publication industry. INCF is represented by national 'nodes' which receive funding from the member states. The workshop participants appreciated Jan Bjaale offering the support of INCF to possible projects in SB applied to research in brain disorders.

Discussion and Results

The purpose of the meeting was to evaluate the feasibility of a systems biology (SB) approach to BGDs by exploring the possibilities of the current technology (functional genomics, imaging, in vitro and in vivo physiology etc.), the availability of existing animal models, the access to patients, and the existing modelling efforts.

Based on this evaluation, the participants discussed and eventually agreed on a ranking of

disorders suitable for a systems biology approach:

Disease	Significance	Data situation	Modelling
#1 Drug addiction	multiple kinds of addiction with common core mechanism; high social impact; targeted treatment required, some clinical ties	good, animal models, large functional genomics projects	common core with neuro adaptation, few quant. models available
#2 Parkinson's disease	1% of 60+ and 5% of 80+ affected locomot vs. psychosis, potential clinical ties	strong, good model systems available, many levels accessible, large proteomics networks	ongoing at many levels: metabolic models, dopamin pathway, neurons, MSN models
#3 Schizophrenia	Very high, possible links to smoking/alcohol addiction, good access to patients	comparable to drug addiction, few and poor model systems	

However, it was generally agreed that the understanding of schizophrenia is still embryonic, and a systematic and systemic approach was maybe premature. Therefore, the panel suggests that effort should be focussed on **Parkinson's Disease** and **Drug Addiction**

Moreover, the participants discussed in detail the situation of experimental data of all levels with regard to availability, quality and value to SB. The results are concisely summarised in the following table:

Type	Specifications	Quality of curr. data	Remarks
<i>Molecular</i>			
Gene expression	- which - time - location - data source	B C B B	
Protein	- which - time - location - data source - interaction - dynamics of i. - modification - structure/function	A-C A-C B-C B-C B B-C B A-C	large-scale data should be made available for 3 neuronal cells: dopaminergic (DA) neurons, striatal medium spiny neurons (MSN) D1 and D2
Lipids		C	
Metabolome		C	
Functional	- kinetics - interactions	C B-C	functional data are largely missing

	- diffusion constants - cell physiology	C C	
<i>Cell</i>			
Morphology		A-B	
Electrophysiol.	- passive (Rm, Cm, Vm, Ra) - channel (kinetics, distribution) - transmitter - firing patterns - plasticity - shape (changes)	A-B B-C A A A-C B-C	
<i>Circuitry</i>			
Connectivity	- statistics - location - type (chem/gap)	B-C B-C B-C	systematic microscopy of neuron morphology and brain structures available for reconstruction
Activity	- unit - field - EEG - fMRI (patients)	B B A	
Structure	- MRI - spectroscopy	A A	
<i>Behaviour</i>			
Disease models		?	
Patients		?	

Considering what is necessary, what is available, where standardisation is mature, and where experimenter's expertise is still the main factor, the panel concluded that a call for funding Systems Biology approaches to basal ganglia should also fund quantitative data gathering along two different lines:

- Large-scale data gathering, centralised in a few experimental facilities should seek to gather information about the protein content of three major cells: mesencephalic dopamine neuron, striato-nigral ("D1") medium-spiny neuron and striato-palidal ("D2") medium-spiny neuron. These large-scale effort should determine at least: the proteome, the interactome, the concentration of the proteins and their subcellular locations. Standardisation and public databases are ready.
- Kinetics information gathering should be funding using classical distributed funding. This information concerns chemical kinetics constants, diffusion constants, electrophysiological parameters of subcellular compartments, dynamics of neurotransmitter release.

The requirements of modelling with regard to computing aspects (standardisation, access, etc.) and options for collaboration shall be discussed separately at the upcoming workshop "Dynamic Principles of Cellular Function" on October 24th, 2007 in Frankfurt/M, Germany.