Epilepsy Genetics Workshop & Young Researchers in Epileptology Meeting 2013

Dec 12th – 14th, 2013
Beer Sheva & Sde Boker
Israel

PROGRAM & ABSTRACTS
Dear Colleagues and Friends,

We are happy to welcome you to the

Sde Boker – Meeting on Epilepsy Genetics and the Young Researchers in Epileptology – Meeting 2013

taking place in Midreshet Sde Boker, Ben-Gurion University Campus for Desert Research (Israel) from December 12th to December 14th, 2013.

Within this beautiful part of the Negev desert, the home of Israel’s first prime minister David Ben Gurion, we will find an inspiring atmosphere to discuss the latest developments in epilepsy genetics with specific respect to the chances and possibilities of epilepsy genetics in Israel and Palestine. Leading experts in the field will provide an overview of the most recent topics in epilepsy genetics and colleagues from Israel and Palestine will give insight into the specific situation in their countries.

On December 14th, 2013, we are looking forward to continue the success and fun of the Young Researchers in Epileptology-Meeting that was held for the first time in Kiel, Germany, in August 2012. Young scientists and clinicians will get to know each other, present their work and learn from each other as well as from experienced researchers in the field of epilepsy genetics.

Welcome to Israel - have a great time in Sde Boker!

Sarah von Spiczak, Lior Greenbaum, Ingo Helbig, Zaid Afawi, Alon Friedman, Ilan Blatt and Amos Korczyn
This meeting has been made possible by the financial support of:

the Department of Neuropediatrics at the University Medical Center Schleswig-Holstein, Campus Kiel
and the Zlotowski Center for Neuroscience, Ben-Gurion University of the Negev.

In addition, travel support for young researchers was granted by the German Epilepsy Society (DGfE)
and the Israeli Chapter of the International League Against Epilepsy conceded financial support.

The European Science Foundation (ESF) supports the meeting as part of the EuroEPINOMICS dissemination program.

Additional support has been granted by

Prof. Dr. Peter und Jytte Wolf - Stiftung für Epilepsie

http://www.epilepsiestiftung-wolf.de/

as well as by

We are very grateful to all sponsors.
Meeting Venue:

Thursday, Dec 12th, 2013
Senate Building, BGU
(see the attached map)

Friday, Dec 13th, 2013
Conference hall

Dinner Venues:

Thursday, Dec 12th, 2013
Lakia

Friday, Dec 13th, 2013
Sde Boker Dining Room

Saturday, Dec 14th, 2013
Mizpe-ramon / barbecue
BGU campus map

Senat Building 71

Train Station
# PROGRAM

**Thursday Dec 12th**

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
<tbody>
<tr>
<td>8:30 AM</td>
<td>Registration</td>
</tr>
</tbody>
</table>
| 9:00 – 9:30 AM | Welcome to Be’er Sheva  
|             | Amos Korczyn, Ingo Helbig, Gabriel Schreiber, Dean, Faculty of Health Sciences |

### Session 1 – Epilepsy & Epilepsy Genetics in the Middle East  
(Chairs: Amos Korczyn, Hussam Al-Amle)

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
</table>
| 9:30 AM – 1:30 PM | Epilepsy in Israel  
|             | Ilan Blatt (Tel Aviv, Israel)                                          |
| 09:50 – 10:10 AM | Epilepsy and neuropediatrics in Palestine  
|             | Reinhard Keimer (Bethlehem, Palestine)                                 |
| 10:10 – 10:40 AM | The Israel Epilepsy Family Project – the 1st decade  
|             | Samuel F. Berkovic (Melbourne, Australia)                              |
| 10:40 – 11:10 AM | The Israel Epilepsy Family Project – moving ahead  
|             | Karl-Martin Klein (Marburg, Germany)                                   |
| 11:10 – 12:10 AM | Epilepsy genetics in Israel and Palestine— the recruitment pipeline  
|             | Zaid Afawi (Be’er Sheva, Israel)                                       |
| 12:10 – 12:30 AM | Panel discussion                                                        |
| 12:30 AM – 1:30 PM | Lunch                                                                      |

### Session 2 – The big picture in epilepsy genetics  
(Chairs: Bruria Ben Zeev, Frederick Andermann)

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
</table>
| 1:30 – 4:00 PM | The genetics of idiopathic generalized epilepsies  
|             | Holger Lerche (Tubingen, Germany)                                    |
| 2:00 – 2:20 PM | The genetics of idiopathic focal epilepsies with rolandic spikes  
|             | Johannes Lemke (Bern, Switzerland)                                    |
| 2:30 – 2:50 PM | The genetics of rare epilepsy syndromes  
|             | Sarah Weckhuysen (Antwerp, Belgium)                                   |
| 3:00 – 3:20 PM | The genetics of syndromal and metabolic epilepsies  
|             | Tally Lerman-Sagie (Holon, Israel)                                    |
| 3:30– 4:00PM | Coffee break                                                          |

### Session 3 – More than just sequence: epigenetics in epilepsy  
(Chairs: Holger Lerche, Ilan Blatt)

<table>
<thead>
<tr>
<th>Time</th>
<th>Event</th>
</tr>
</thead>
</table>
| 4:00 – 5:30 PM | Epigenetics: an introduction  
|             | Ramon Birnbaum (Be’er Sheva, Israel)                                 |
| 4:30 – 4:50 PM | Methylation and histone acetylation: epigenetic mechanisms in epilepsy  
|             | Katja Kobow (Erlangen, Germany)                                      |
| 5:00 – 5:20 PM | Gene regulation and epilepsy: the role of microRNAs  
|             | Noam Shimron (Tel Aviv, Israel)                                      |
| 5:30 PM – | Dinner in the Bedouin village Lakia  
|             | Greetings-  
|             | Rivka Carmi, President, Ben-Gurion University of the Negev            |
|             | Moving to Sde Boker                                                  |
### Friday, Dec 13th

**Session 4 – Methods and techniques, a hand-on session**  
(Chairs: Bobby Koeleman, Dennis Dlugos)

<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Speaker and Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>9:00 AM – 2:00 PM</td>
<td>The apple and the tree – linkage analysis</td>
<td>Bobby Koeleman (Utrecht, Netherlands)</td>
</tr>
<tr>
<td>9:00 – 9:30 AM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>10:45 – 11:15 AM</td>
<td>G-WHAT? – Genome-wide association studies</td>
<td>Lior Greenbaum (Sheba, Israel)</td>
</tr>
<tr>
<td>11:30 – 12:00 AM</td>
<td>Missing bits and pieces – CNV analysis</td>
<td>Dennis Lal (Cologne, Germany)</td>
</tr>
<tr>
<td>12:15 – 12:45 AM</td>
<td>BAMD, here is your exome – NGS analysis</td>
<td>Arvid Suls (Antwerp, Belgium)</td>
</tr>
<tr>
<td>1:00–2:00 PM</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Impulse:**  
(Chair: Samuel F. Berkovic)

<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Speaker and Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>2:00 – 2:30 PM</td>
<td>From science to clinic: will research in epilepsy genetics revolutionize patient care and treatment?</td>
<td>Ulrich Stephani (Kiel, Germany)</td>
</tr>
</tbody>
</table>

**Session 5 – Genetics and beyond**  
(Chairs: Ulrich Stephani, Alon Friedman)

<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Speaker and Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>2:45 – 4:00 PM</td>
<td>Genetic imaging</td>
<td>Michael Siniatchkin (Frankfurt, Germany)</td>
</tr>
<tr>
<td>2:45 – 3:05 PM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>3:15 – 3:35 PM</td>
<td>Genetics and imaging of the blood-brain barrier</td>
<td>Alon Friedman (Be’er Sheva, Israel)</td>
</tr>
<tr>
<td>3:45 – 4:05 PM</td>
<td>A common SCN1A splice site polymorphism modifies the effect of carbamazepine on cortical excitability - a pharmacogenetic TMS-study</td>
<td>Felix Rosenow (Marburg, Germany)</td>
</tr>
<tr>
<td>4:15 – 4:45 PM</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Session 6 – The new borderland**  
(Chairs: Michael Siniatchkin, Eva Andermann)

<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
<th>Speaker and Location</th>
</tr>
</thead>
<tbody>
<tr>
<td>4:45 – 7:00PM</td>
<td>Genetics of autism spectrum disorder</td>
<td>Sagiv Shifman (Jerusalem, Israel)</td>
</tr>
<tr>
<td>4:45 – 5:05PM</td>
<td></td>
<td></td>
</tr>
<tr>
<td>5:15 – 5:35 PM</td>
<td>Autism and epilepsy: The clinical and neurobiological phenotype derived from Autism-Spectrum Disorder research</td>
<td>Christine Freitag (Frankfurt, Germany)</td>
</tr>
<tr>
<td>5:45 – 6:05PM</td>
<td>Connectivity in autism and epilepsy</td>
<td>Ilan Dienstein (Be’er Sheva, Israel)</td>
</tr>
<tr>
<td>6:15 – 6:35PM</td>
<td>The epilepsy-autism overlap – joining forces?</td>
<td>Yoav Kohn (Jerusalem, Israel)</td>
</tr>
<tr>
<td>6:45 – 7:00PM</td>
<td></td>
<td>Panel discussion</td>
</tr>
<tr>
<td>7:00PM –</td>
<td></td>
<td>Dinner</td>
</tr>
</tbody>
</table>
Saturday Dec 14th  

**Young Researchers in Epileptology**

**Session 7: Young Researchers Meeting**

*(Chairs: Lior Greenbaum, Sarah von Spiczak)*

**Part A – Presentation of Projects: A brain storming session**

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
</tr>
</thead>
<tbody>
<tr>
<td>8:30 AM – 1:00 PM</td>
<td><strong>Introduction</strong>&lt;br&gt;<strong>Lior Greenbaum, Sarah v. Spiczak</strong></td>
</tr>
<tr>
<td>8:30 – 8:45 AM</td>
<td><strong>A life-long career in epilepsy genetics: what can we learn from the experienced?</strong>&lt;br&gt;<em>Eva Andermann (Montreal, Canada)</em></td>
</tr>
<tr>
<td>8:45 – 9:15 AM</td>
<td><strong>Presentations by Young Researchers: First round</strong>&lt;br&gt;(short introductory talks, 5 min/5 slides, discussion at posters)</td>
</tr>
<tr>
<td>9:15 – 10:45 AM</td>
<td><strong>Presentations by Young Researchers: Second round</strong>&lt;br&gt;(short introductory talks, 5 min/5 slides, discussion at posters)</td>
</tr>
<tr>
<td>10:45 – 11:00 AM</td>
<td>Coffee break</td>
</tr>
<tr>
<td>11:00 – 12:30 AM</td>
<td><strong>Lunch</strong></td>
</tr>
</tbody>
</table>

---

**Part B – Group workshops: So, you want to be a scientist?**

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
</tr>
</thead>
<tbody>
<tr>
<td>1:30 – 5:00 PM</td>
<td><strong>חוצפה, zen and Fahrvergnügen – a motivational talk</strong>&lt;br&gt;<em>Ingo Helbig (Kiel, Germany)</em></td>
</tr>
<tr>
<td>1:30 – 2:00 PM</td>
<td><strong>Plan, perform, publish: how to set up a project in epilepsy research</strong>&lt;br&gt;<strong>Round table: Young Researchers asking the experienced:</strong>&lt;br&gt;- How to plan your project&lt;br&gt;- How to deal with ethical aspects&lt;br&gt;- How to finance your project&lt;br&gt;- How to find partners for your project&lt;br&gt;- How to publish your project&lt;br&gt;- ...</td>
</tr>
<tr>
<td>2:00 – 3:30 PM</td>
<td>Coffee Break</td>
</tr>
</tbody>
</table>

---

**Part C – Back to the bedside: The big goal of epilepsy research**

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
</tr>
</thead>
<tbody>
<tr>
<td>4:30 – 5:45 PM</td>
<td><strong>Discover new genes in epilepsy: what do we need?</strong>&lt;br&gt;<em>Orly Elpeleg (Jerusalem, Israel)</em></td>
</tr>
<tr>
<td>4:30 – 5:00 PM</td>
<td><strong>Lost in translation</strong>&lt;br&gt;<em>Amos Korczyn (Tel Aviv, Israel)</em></td>
</tr>
<tr>
<td>5:00 – 5:30 PM</td>
<td><strong>Challenges of genomic applications in healthcare systems</strong>&lt;br&gt;<em>Moein Kanaan (Bethlehem, Palestine)</em></td>
</tr>
<tr>
<td>5:30 – 6:00 PM</td>
<td><strong>Closing remarks</strong></td>
</tr>
<tr>
<td>6:00 – 6:15 PM</td>
<td><strong>Dinner in the desert</strong></td>
</tr>
</tbody>
</table>
ABSTRACTS

A: Invited talks (abstracts 1-3)
B: Young researchers (abstracts 4-15)
Abstract
Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name Sam Berkovic (on behalf of a large collaborative group)

Title The Israel Epilepsy Family Project – the 1st decade

**Background:** Since 1998 we utilized the unique population of Israel, with high level medical services and a very cooperative neurology community to study epilepsy genetics. In order to enrich for genetic causation, but to avoid selecting only large families with obvious Mendelian segregation, we studied multiplex families. Families were evaluated to determine the electro-clinical phenotype, inheritance patterns and molecular lesions.

**Methods:** We sought families with two or more subjects with epilepsy from clinicians throughout Israel. Following referral, individuals were diagnosed with epilepsy syndromes, each of which were assigned into the broad categories of generalized, focal, GEFS+ and special syndromes. Classification of families into these same categories were made following the diagnosis of at least two family members; families were designated ‘mixed” when more than one syndrome was diagnosed in different family members. Pedigrees were analysed, ethnicity was recorded and molecular genetic studies were performed.

**Results:** 211 families met our inclusion criteria. We successfully classified 169 families (80%) into broad familial epilepsy syndrome groups: 69 Generalized, 22 Focal, 24 GEFS+, 32 Special, and 29 Mixed syndromes. 42 families remained unclassified. Arab families made up 25% of our cohort with the remaining families Jewish (44% Sephardic, 23% Ashkenazi, 8% mixed Jewish). Arab families were disproportionately represented in our Special familial syndrome group and were more likely to be consanguineous.

Molecular lesions were identified in 47/211 families. The majority of these findings were made in established epilepsy genes (e.g., SCN1A, KCNQ2, SLC2A1, CSTB) chosen for investigation following careful clinical characterization, although the discovery of a dominant SCN1A mutation in a family with severe focal epilepsy was a surprise. Detailed clinical and molecular studies in this cohort, contributed to earlier reported original genetic discoveries in ten families (e.g., KCNT1, PCDH19, TBC1D24). The independent discovery of LAMC3 in a large consanguineous family was unexpected. The predominant familial phenotype was epilepsy with myoclonic atonic seizures, however, subsequent brain imaging confirmed an occipital dysplasia consistent with the single previous report.

**Conclusion:** Causative mutations were identified in no less than 22% of the total cohort. Clinical and molecular genetic findings were made that are both unique to the Israeli cohort but others representative of the wider population highlighting the value of genetic studies in distinctive populations.
Abstract
Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

<table>
<thead>
<tr>
<th>Name</th>
<th>Ramon Birnbaum</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>The Life Science Department, Ben Gurion University, Be’er Sheva, Israel</td>
</tr>
</tbody>
</table>

| Title                        | Functional genomic characterization of gene regulatory elements associated with epilepsy |

Epilepsy is a complex and heterogeneous disease making it difficult to precisely diagnose and provide effective treatments. A major and underexplored cause of complex disorders such as epilepsy could be mutations in gene regulatory elements, such as enhancers. To test this hypothesis, we decided to focus on infantile spasms (IS), a subtype of epilepsy which begins in infancy and is associated with ventral forebrain development and forebrain synapse function. Using chromatin immunoprecipitation followed by deep sequencing (ChIP-seq) with enhancer marks (H3K27ac, RNAPoll2), we identified active enhancer candidates in mouse embryonic day 16.5 forebrain. In addition, using chromatin interaction analysis followed by paired-end tag sequencing (ChIA-PET) on the same tissue, we determined the enhancer candidates that regulate IS-associated genes. Using zebrafish transgenic assays, we show that several of these candidates have enhancer activity in the developing forebrain. Our results provide a novel dataset of neuronal enhancers that are involved in the spatiotemporal regulation of IS-associated genes. In addition, this work will shed light on neuronal gene regulation in general and identify novel genomic regions that could be involved in epilepsy pathogenesis and brain development.
Abstract

Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name
Katja Kobow
Institute of Neuropathology, University Hospital Erlangen, Erlangen, Germany

Title
Epigenetic mechanisms in epilepsy

Epigenetic mechanisms are self-perpetuating, posttranslational modifications of nuclear proteins (acetylation, methylation, phosphorylation, etc.), and DNA (methylation) that can produce lasting alterations in chromatin structure and gene expression patterns. They are increasingly recognized as fundamental regulatory processes in central nervous system development, synaptic plasticity, and memory. Epigenetic alterations are implicated in many neurological disorders including autism, bipolar disorders, schizophrenia, brain tumors, neurodegeneration, and more recently epilepsy.

We used a massive parallel sequencing approach to map genome-wide alterations in DNA methylation in a chronic rat TLE model. Sequencing of mRNA was used in same specimens for complementary gene expression profiling and integration with methylome data. Unsupervised clustering of an epigenetic mark, i.e. DNA methylation, successfully separated chronic epileptic from non-epileptic animals (Kobow et al., 2013). Further, aberrant methylation patterns could be inversely correlated with gene expression changes. Complementary data on altered microRNA, histone modification and DNA methylation dynamics in experimental and human epilepsy further support our idea that epigenetic gene regulation may be critical in epileptogenesis and propagation of the chronic disease state. We suggest that addressing epigenetic mechanisms may be a successful strategy to increase the brain’s sensitivity to antiepileptic drugs and may even act as disease-modifying treatment.
Abstract

Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name
Ann-Kathrin Ruppert
Cologne Center for Genomics, University of Cologne, Cologne, Germany

Title
Parent-of-origin effects in genetic generalized epilepsies
A-K Ruppert, H Schulz, T Sander

Genetic generalized epilepsies (GGEs) affect about 0.3% of the general population and account for 30% of all epilepsies. Heritability estimates of about 80% suggest a predominant genetic etiology of common GGE syndromes. Epigenetic regulatory mechanisms, such as DNA methylation, histone code modifications and chromatin remodelling, have been implicated in epileptogenesis (review in: Qureshi & Mehler. Neurobiol Dis 2010;39:53-60). Evidence for a preponderance of maternal inheritance and a striking female excess implicates the involvement of epigenetic effects in the pathogenesis of GGE (Pal et al. Brain Dev 2006;28:92-8).

Parent-of-origin (PofO) effects, such as imprinting are a phenomenon in which homologous chromosomes exhibit differential gene expression and epigenetic modifications according to their parental origin (Garg et al. PLoS One 2012;7:e41695). Such non-Mendelian inheritance patterns are generally ignored by conventional association studies, as these tests consider the maternal and paternal alleles as equivalent. To identify potential PofO effects we associate SNP genotypes of defined parental origin with methylation levels: first of all a dissection of differentially methylated parental genomic regions (imprinting mQTLs: imQTLs) was made. The screening for imQTLs was performed by comparing CpG methylation states of GGE trio offsprings with reciprocal heterozygote SNP genotypes of inverse parental origin (A\texttt{MAT}B\texttt{PAT} versus A\texttt{PAT}B\texttt{MAT}) (Garg et al. PLoS One 2012;7:e41695). Our screening method revealed an enrichment of imQTLs within known imprinted regions. This observation demonstrates the validity of this approach to detect known and novel genomic regions that differentially influence CpG methylation and gene expression in a PofO specific manner. Subsequently, PofO association analysis was carried out in an assembly of imprinted genomic regions in the methylome of blood cells in 566 parent-offspring GGE trios. Considering that only one parental allele is expressed at imprinted loci, the active parental allele has a stronger impact on the gene function compared with alleles displaying a diploid gene expression. Therefore, susceptibility alleles conferring a PofO effect should be enriched in imprinted regions and the focus on a smaller part of the genome increases the analytical power.

In conclusion, combining of imQTL and PofO association analysis has the potential to increase the analytical power to detect susceptibility factors with PofO effects which might not be detected by an unspecified GWAS.
Abstract
Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name | Christina Selch
Schön Klinik Vogtareuth, Germany

Title | Therapy of Dystonia and Epilepsy in 5 Patients with ARX-mutations

**Objectives:** Mutations in the X-chromosomal ARX (Aristaless-Related-Homeobox) gene cause both nonsyndromic and several forms of syndromic mental retardation. The phenotypes associated with mutations of ARX are characterized by pleiotropy. Common clinical features include epilepsy, brain malformations, genital abnormalities and dystonia. Dystonia has previously described as a clinical feature in patients with ARX mutations, however, there is hardly any information about antidystonic treatment in affected patients.

**Methods:** Retrospective evaluation of 5 patients (all boys, age 4-17). All patients had epilepsy, 3/5 had dystonia.

**Results:** All patients had infantile spasms with onset at 4-6 months and further developed generalized tonic-clonic seizures. Epilepsy was treated with multiple antiepileptic drugs including benzodiazepines, barbiturates, steroids, valproic acid, vigabatrin, levetiracetam and oxcarbazepine. All patients responded to combination therapy including VPA, 2 became seizure free. Dystonia in all 3 patients presented in the first months of life, one had severe status dystonicus at the age of 10 years. Dystonia was successfully treated with tetrabenazine (1/3), oral baclofen (3/3), intrathecal baclofen (1/3) and L-DOPA (1/3).

**Conclusion:** Besides the epilepsy, dystonia is an important clinical feature in some patients with ARX mutations. Dystonic symptoms may present even before the onset of epilepsy in early infancy and may be difficult to differentiate from epileptic seizures. Successful therapeutic options include oral and intrathecal baclofen. No general conclusions can be drawn from this case evaluation but may give an impulse for collaborated clinical observations and data collections.
Abstract
Sde Boker – Meeting on Epilepsy Genetics
&
Young Researchers in Epileptology – Meeting 2013

Name
Eva Maria Reinthaler
Department of Neurology, Medical University of Vienna, Vienna, Austria

Title
Recurrent genomic rearrangement confers risk for typical and atypical Rolandic Epilepsy
(* contributed equally)

Recurrent microduplications and microdeletions have been implicated in a variety of childhood-neurodevelopmental diseases. We demonstrate that one of these genomic rearrangements is associated with typical and atypical Rolandic epilepsy (number of chromosome and precise position will be announced at the poster). Rolandic Epilepsy is the most common idiopathic focal childhood epilepsy and its genetic basis is largely unknown. In a cohort of 393 patients, we identified five patients (1.27%) carrying this recurrent copy number variant and a sixth patient (1.53% in total) with an atypical shorter rearrangement. Compared with 1/3768 (0.03%) controls the rearrangement is significantly associated with typical and atypical Rolandic Epilepsy (Fisher’s exact test, \( P = 4.4 \times 10^{-6} \), OR = 58.3, 95% CI: 7.04-2643.11). The copy number variant was not detected in 350 patients with mesial temporal lobe epilepsy, an adult form of focal epilepsy. Our results further support the pathogenicity of recurrent copy number variants in early development and expand the range of phenotypes to include typical and atypical Rolandic Epilepsy.

EuroEPINOMICS Consortium contributing partners:
Martha Feucht, Hannelore Steinböck, Birgit Neophytou, Gabriel M. Ronen, Laurian Roche, Ursula Gruber-Sedlmayr, Julia Geldner, Edda Haberlandt
Abstract
Sde Boker – Meeting on Epilepsy Genetics
&
Young Researchers in Epileptology – Meeting 2013

Name
Florans Madjidyar
Department of Neuropediatrics, University Medical Center Schleswig-Holstein, Kiel, Germany

Title
Prospective screening of GLUT1-defects on pediatric patients detecting prevalence and phenotypic spectrum

Introduction: Glucose transport protein type 1 (GLUT1) facilitates the transport of glucose from the bloodstream across the blood-brain barrier to the central nervous system. GLUT1 is encoded by the SLC2A1 gene; mutations occurring de novo or being inherited result in cerebral energy failure and seizures.

Patients with the "classical" form of GLUT1 deficiency syndrome present with infantile seizures, developmental delay, acquired microcephaly, hypotonia, spasticity, and a complex movement disorder consisting of ataxia and dystonia. The phenotypic spectrum expands every year and a variability of signs and symptoms were recognized. Beside severe epileptic encephalopathy’s accompanied by dystonia and mental retardation, mutations were found in patients with carbohydrate-responsive symptoms, with predominant ataxia or dystonia but without seizures, and with paroxysmal exertion-induced dyskinesia and seizures.

Early diagnosis of SLC2A1-mutations is critical because it allows prompt initiation of treatment with a ketogenic diet and shows improvement of seizure and movement disorders.

Methods: We prospectively recruited 65 consecutive pediatric patients with newly diagnosed epilepsy and/or at their first visit in our center from July 2012 until March 2013. The cohort included patients having idiopathic generalized epilepsy as MAE (myoclonic astatic epilepsy) and EOAE (early onset absence epilepsy), idiopathic focal epilepsy, febrile seizures and other epileptic syndromes. In addition, a population-based cohort “popgen” of 206 patients was investigated, including similar epileptic syndromes as aforementioned. The coding regions of the SLC2A1-gene were analyzed by Sanger-sequencing.

Results: Apart from single nucleotide polymorphisms, no mutations were found.

Conclusion: Mutations in SLC2A1 are rare in a mixed cohort of pediatric patients with various epilepsy syndromes.
Abstract
Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name
Gali Heimer
Pediatric neurology unit, Edmond and Lily Safra Children Hospital, Sheba Medical Center, Israel

Title
Glycosylphosphatidylinositol (GPI) anchor proteins defects – a new important cause for early infantile epileptic encephalopathy
G Heimer, B Pode-Shaked, D Marek-Yagel, M Tzadok, E Genelin, A Nissenkorn, Y Anikster, B Ben-Zeev

Over the last few years, mutations in eight genes involved in the biosynthesis of glycosylphosphatidylinositol (GPI) anchor proteins have been reported. Up to now disease causing mutations have been described in PIGA, PIGL, PIGM, PIGN, PIGO, PIGV, PGAP2 and PIGT. These constitute a new subclass of congenital disorders of glycosylation and although each differs in its distinct set of clinical features and multi-organ involvement they all have in common central hypotonia, intellectual disability (ID) and epilepsy.

We report here a series of four patients from three families with mutations in three different GPI anchor proteins, all causing epilepsy as one of their core clinical features and in three of them in the form of early infantile epileptic encephalopathy (EIEE).

Patient I - a girl of mixed Ashkenazi-Moroccan descent, presented with myoclonic seizures from birth with evolution of hypersynchronous pattern, severe ID, hypotonia, cortical blindness, cardiomyopathy and vesico-urethral-reflux with hydronephrosis. Whole exome sequencing in a "trio" format was applied and revealed a novel missense mutation in the PIGN gene. Patient II - a girl of an Arabic origin presented with mild-moderate ID, portal vein thrombosis and an atypical absence epilepsy that manifested at the age of 3 years. The girl has six siblings, two of whom died from the same disease. The unique presentation led to a 'spot diagnosis' and sequencing revealed a mutation in the promoter of the PIGM gene. Patients III,IV are two brothers of a Yamani descent that presented with EIEE with mixed seizure types that appeared before the age of one month, severe ID, stereotypic movements, microcephaly, facial dysmorphism and central hypotonia with peripheral spasticity. A novel missense mutation in the X-linked PIGA gene was found using whole exome sequencing, also in a "trio" format.

It is noteworthy that in all patients epilepsy was a major feature and moreover in three of them it was consistent with the definition of EIEE. It is also noteworthy that none of these patients was found to have primary developmental cortical malformation or migration defect suggesting their epilepsy is a direct consequence of the metabolic pathways influenced by the defects in the GPI anchor proteins.

We propose that mutations in GPI anchor proteins have a primary epileptogenic effect and should be considered in children of all origins with EIEE accompanied with hypotonia, intellectual disability with or without additional multi-organ involvement.
Abstract
Sde Boker – Meeting on Epilepsy Genetics
&
Young Researchers in Epileptology – Meeting 2013

Name
Hille Harms
Department of Neuropediatrics, University Medical Center Schleswig-Holstein, Kiel, Germany

Title
Exome sequencing in recessive familial epilepsies – identification of novel epilepsy genes
H Harms, M Pendziwiat, S Appenzeller, J Jähn, S von Spiczak, H Muhle, Z Afawi, U Stephani, G Kuhlenbäumer and I Helbig

Introduction: In families with two or more affected children and unaffected parents, a recessive mode of inheritance is usually assumed plausible. However, known metabolic, neurodegenerative or mitochondrial disorders are only present in a very small subset of families and a large genetic heterogeneity is assumed in familial seizure disorders. Novel high-throughput technologies now allow for massive parallel sequencing to identify possible candidate genes on an exome-wide basis.

Methods: 4 families with recessive families were included in a pioneer project to assess the feasibility of a novel bioinformatics pipeline for variant analysis. Whole exome sequencing was performed followed by variant annotation and filtering for homozygous variants after exclusion of inhouse variants.

Results: Using the technology of whole-exome sequencing in 4 recessive families, we were able to narrow down the possible causative gene to a single variant in 2/4 families. Segregation analysis confirmed heterozygous status in the unaffected parents and – if available – siblings.

Discussion: We were able to demonstrate that the technology of whole exome sequencing allows us to zero in on promising candidate genes in 2/4 families included in our pioneer study using a “single-exome” approach. For families negative for promising candidate genes, a family-based sequencing approach or whole-genome approach may be feasible rather than subsequent validation of numerous candidate variants.
Abstract
Sde Boker – Meeting on Epilepsy Genetics &
Young Researchers in Epileptology – Meeting 2013

Name
Klari Noormets
Children’s Clinic of Tartu University Clinic, Tartu, Estonia

Title
Copy number variants associated with drug-resistant epilepsy in Estonia
K Noormets, I Talvik, T Reimand, E Öiglane-Šlik, K Öunap, T Talvik

Background: Drug-resistant epilepsy is a serious problem for the families and physicians. Several chromosomal abnormalities have been described that cause intractable epilepsy but the role of many copy number variants (CNV) is unknown. Microarray-based genomic copy-number analysis (chromosomal microarray - CMA) gives a chance to detect very small chromosomal imbalances associated with different diseases.

Aim: To find out how many patients with drug-resistant epilepsy in Estonia carry different CNV’s and to determine the relevance of these mutations according to epilepsy.

Patients and methods: Estonian database of CMA analyses consists data of 1100 patients, analyzed since 2009. 165 of them are children with epilepsy aged 0-18 years. From these 165 children 78 (47,3%) meet the criteria of ILAE definition of drug-resistant epilepsy.

Results: CMA changes were found in 46/165 (27,9%) of children with epilepsy. In the study group of 78 children with drug resistant epilepsy CMA changes were found in 31 (39,7%). Of these 31 loss of heterozygosity (LOH) was found in 8 (25,8%), duplications in 10 (32,3%) and deletions in 13 (41,9%) cases. In most cases the association with epilepsy is not clear but there were also common epilepsy causing mutations present.

Conclusions: CMA is an useful diagnostic tool in detection of CNV's in drug-resistant epilepsy. Almost half of the analyzed patients had CNV’s but further identification of variant loci and the genes within them warrant further evaluation.

The study was supported by the EuroEPINOMICS grant SARLA 11091E
Abstract

Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

Name: Mikko Muona
Institute: Institute for Molecular Medicine Finland (FIMM), University of Helsinki, Helsinki, Finland

Title: Deciphering the genetic background in patients with PEHO-like syndrome

PEHO syndrome (progressive encephalopathy with edema, hypsarrhythmia, and optic atrophy) is an infantile onset neurodegenerative disorder. The clinical presentation in Finnish patients, who are homozygous for a founder mutation (unpublished), is relatively uniform and includes hypotonia, infantile spasms with hypsarrhythmia, profound psychomotor retardation, optic atrophy, progressive brain atrophy, which is most prominent in cerebellum, and dysmyelination. A significant number of patients manifest many of these features in the absence of the typical neuroradiological findings, or with no sign of progression. These patients remain without proper diagnosis but are often classified as PEHO-like.

We aim to decipher the genetic basis in patients with PEHO-like syndrome by using exome sequencing. We have selected 29 Finnish, mostly sporadic, PEHO-like patients, who have been excluded for the PEHO founder mutation. Exome sequencing was performed using Illumina HiSeq 2000 platform. We carried out variant filtering using strategies assuming two different patterns of inheritance: recessive and de novo. In the former we selected potentially deleterious variants – either homozygous or compound heterozygous – of essential splice site, nonsense, frameshift, and missense types with a 1000 genomes minor allele frequency below 1%. In the “de novo” analysis we included only heterozygous, potentially damaging variants absent from the control databases. To facilitate identification of de novo variants, we sequenced exomes of parents of six patients without findings in the recessive analysis.

Analysis of the exomes for recessive mutations revealed an affected sibpair with likely pathogenic compound heterozygous mutations in ABAT, previously linked to GABA-transaminase deficiency syndrome. The “de novo” analysis revealed mutations in three genes previously linked to early infantile epileptic encephalopathies. Two male patients had likely pathogenic hemizygous mutations in CDKL5 with capillary sequencing of the patients’ parents showing that the mutations occurred de novo. One patient had a three-amino-acid duplication previously reported as pathogenic in SPTAN1. De novo analysis of six trios identified a novel missense mutation in SCN2A in one patient. Finally, we have also identified potentially pathogenic mutations in genes without a previous connection to encephalopathies.

Our findings imply that “PEHO-like syndrome” is genetically highly heterogeneous and has overlap with early infantile epileptic encephalopathies. A subset of patients had mutations in previously established disease genes, indicating the utility of exome sequencing as a diagnostic tool.
Abstract
Sde Boker – Meeting on Epilepsy Genetics &
Young Researchers in Epileptology – Meeting 2013

Name
Markus von Deimling
Department of Neuropediatrics, University Medical Center Schleswig-Holstein, Kiel, Germany

Title
Gene expression in absence epilepsy

Absence epilepsies, such as childhood absence epilepsy (CAE) and juvenile absence epilepsy (JAE), account for great parts of genetic generalized epilepsies (GGE), formerly named idiopathic generalized epilepsies (IGE) and have a specific age-dependent onset. Unifying electroencephalographic hallmark is a generalized spike and wave activity accompanied by absence seizures. Implicated in the latest classification a genetic background is supposed whereas hardly any susceptibility genes are known. Microdeletion 15q13.3 is the most common risk factor that was found to date. Thus, we performed a three stage analysis design comparing affected children to healthy controls. For each stage a cohort of 20 participants was recruited consisting of 10 cases and 10 controls in order to assess differences in gene expression levels.

**First stage.** Genome-wide gene expression was measured in human lymphocytes using genechip *Human Genome U133 Plus 2.0* (Affymetrix). Data normalization and two-sample t-tests generated a list of all significant differentially expressed genes.

**Second and third stage.** Investigation embraced the top 75 genes from the list generated in stage one and in addition 21 candidate genes from further studies. Differences in gene expression levels were identified by quantitative real time PCR (qRT-PCR) using *TaqMan Low Density Custom Arrays* (Applied Biosystems).

In summary, the goal of investigation of 30 affected versus 30 unaffected participants is to find novel genes involved in GGE possibly altering the gene expression and to provide a reliable biomarker for clinical use.
# Abstract

**Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013**

| Name            | Roni Cohen  
<table>
<thead>
<tr>
<th></th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td>Department</td>
<td>Department of Pediatric Neurology and Epilepsy Center, Schneider Children's Medical Center of Israel, Tel Aviv, Israel</td>
</tr>
</tbody>
</table>

| Title                                                                 | A patient with electrical status epilepticus during slow-wave sleep and a mutation in the gene encoding the Na⁺/H⁺ exchanger NHE6 (SLC9A6) causing Christianson syndrome  
|-----------------------------------------------------------------------|----------------------------------------------------------------------------------|

Electrical status epilepticus during slow-wave sleep (ESES) is an electrographic pattern which is characterized by marked sleep potentiation of epileptiform activity that leads to near-continuous, bilateral spike-wave discharges during non-REM sleep. The ESES pattern may be associated with uncontrolled seizures and neurocognitive regression. The etiology is often unknown, but genetic risk factors have been implicated. We report a patient with ESES and mutation in the gene SLC9A6 encoding NA⁺/H⁺ exchanger NHE6. Mutations in SLC9A6 are associated with Christenson syndrome which presents with X-linked mental retardation, microcephaly, severe intellectual disability, absence of speech, autistic behavior, gait ataxia, early onset of seizures and cerebellar hypoplasia. Our patient has the characteristic findings, intractable epilepsy and ESES with improvement of neurological symptoms and behavior due to periods with seizure control. We would like to delineate the ESES etiology, the SLC9A6 mutation and the clinical symptoms of Christianson syndrome.
Abstract
Sde Boker – Meeting on Epilepsy Genetics & Young Researchers in Epileptology – Meeting 2013

| Name | Stella Põldsepp  
|      | Children’s Clinic of Tartu University Hospital, Tartu, Estonia |
| Title | A boy with CDKL5-related epileptic encephalopathy: diagnostic work-up  
|       | S Põldsepp, K Noormets, U Vahter, K Õunap, T Reimand, T Talvik, I Talvik |

**Background:** Mutations in the X-linked cyclin-dependent kinase-like 5 (CDKL5) gene are responsible for an early infantile epileptic encephalopathy, characterized by intractable epilepsy, severe mental retardation, and later on the development of Rett syndrome–like features. Most of the CDKL5 mutations are identified in girls and only a few mutations are found in boys.

**Clinical case:** A boy started to have seizures at the age of 1.5Mo. At 2.5Mo he had generalized clonic seizures. EEG showed generalized epileptiform discharges, MRI was normal and treatment with valproic acid was initiated. At the age of 3Mo he presented series of spasms and myoclonias, neurological examination showed poor psychomotor development: lack of eye contact, no head control and central hypotonia. EEG revealed atypical hypersarrhythmia and West syndrome was diagnosed. He was treated with glucocorticoids (pulse therapy with i/v methyl-prednisone and oral prednisone) along with vigabatrin. After being 3 weeks seizure-free, epileptic spasms and myoclonias recurred. Extensive metabolic work-up was negative. At 5Mo sequencing of CDKL5 revealed a heterozygous mutation (c.2225_2228, p.E742Afs*41) and CDKL5-related epileptic encephalopathy was diagnosed. At the age of 8Mo there is no positive dynamics of psychomotor development: still no eye contact and head control and severe central hypotonia is persisting. He has several myoclonias during the day and EEG reveals multifocal interictal epileptiform discharges awake and burst-suppression like pattern in sleep. Inspite of AED polytherapy, he has still seizures.

**Conclusion:** This is the second case of CDKL5-related epileptic encephalopathy in Estonia. Genetic testing of CDKL5 mutation should be considered with severe early-onset epileptic encephalopathy.

The research was supported by the EuroEPINOMICS grant SARLA 11091E.
The most beautiful thing we can experience is the mysterious. It is the source of all true art and science.

Albert Einstein