Using the zebrafish to understand and develop treatments for complex brain disorders

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SLC6A1 Round Table
American Epilepsy Society Meeting
New Orleans, USA
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CONFIDENTIAL

Chemical Neuroscience Group

What is our research about?

- *Study the causes of brain disorders*
  - ***Epilepsy**, Schizophrenia/Bipolar, Autism*
- *Create new animal models of human disease*
- *Use models to screen for novel drugs*
- *Understand how these drugs work*
- *Test drug candidates for potential toxicities*

What is our long-term goal?

- *Develop new, safe and effective medicines*

Zebrafish: a novel way of "fishing"

- Vertebrate model with fully sequenced, annotated genome
- Strong genetic, physiological and pharmacological similarity to humans
- High fecundity and small size (96-well format)
- Rapid development *ex utero*
- Optical transparency (non-invasive imaging)
- Only μg amounts of compounds needed; readily absorbed (skin, GI tract, gills)

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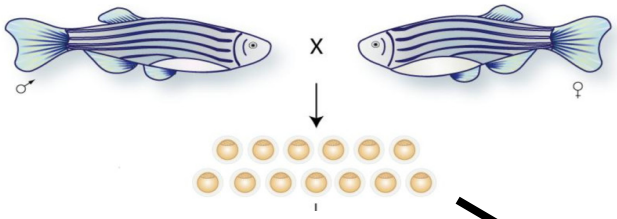
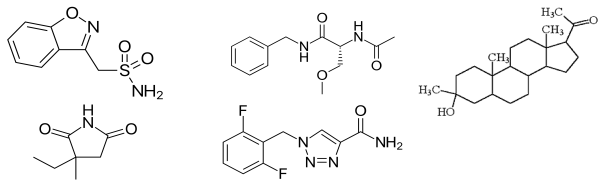
Chemical Neuroscience Group Platform

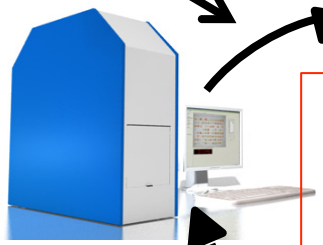
Mutants



Compound libraries

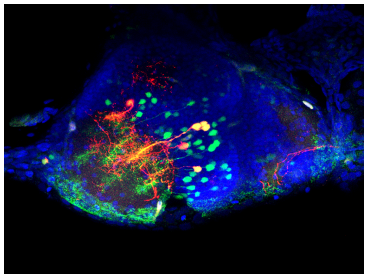


Phenotypic Analysis

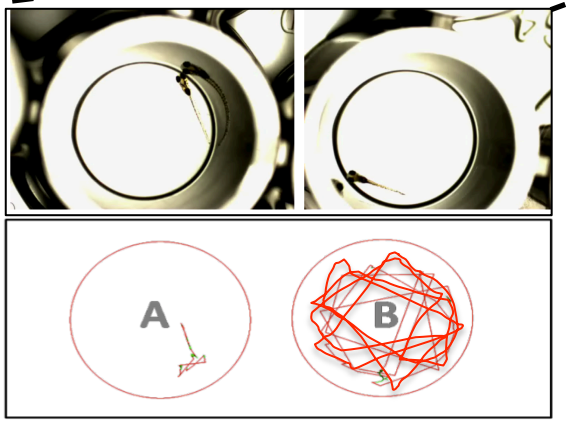


***in vivo* drug screen**

Pharmacology, Toxicology
Histology, RNA-Seq, Imaging
Neurotransmitter Profiling

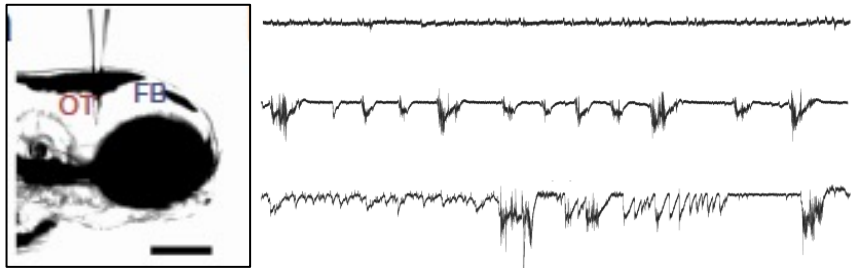


Behavioral assays



wild-type mutant

EEG confirmation of seizure inhibition



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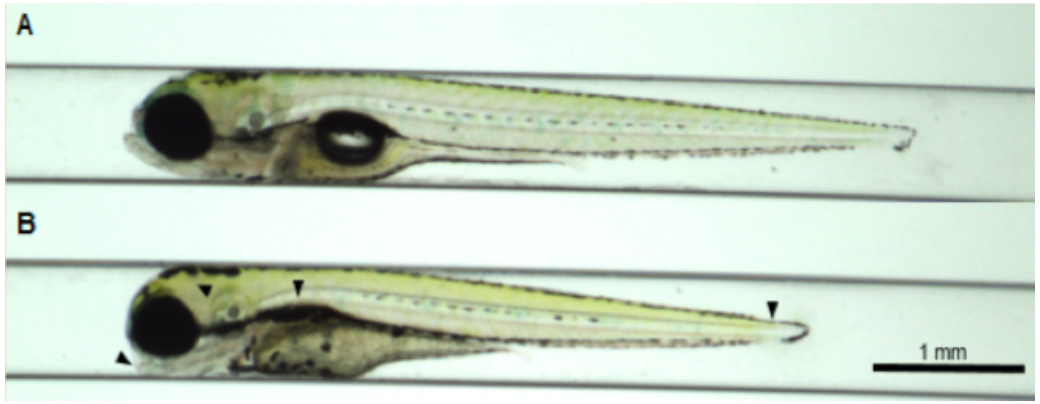
Genetic zebrafish mutant for severe early onset epilepsy syndrome



5 days post fertilization wildtype (A) and mutant (B)

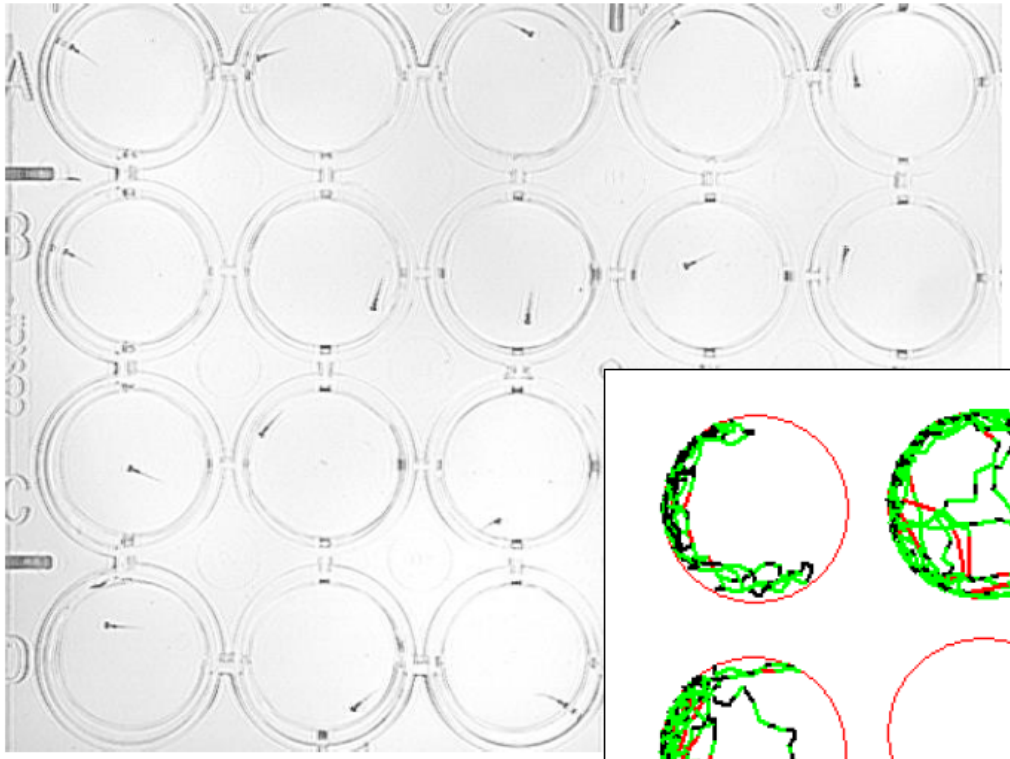
Patients:

- Physically underdeveloped
- Slow movements
- Seizures
- Autistic features

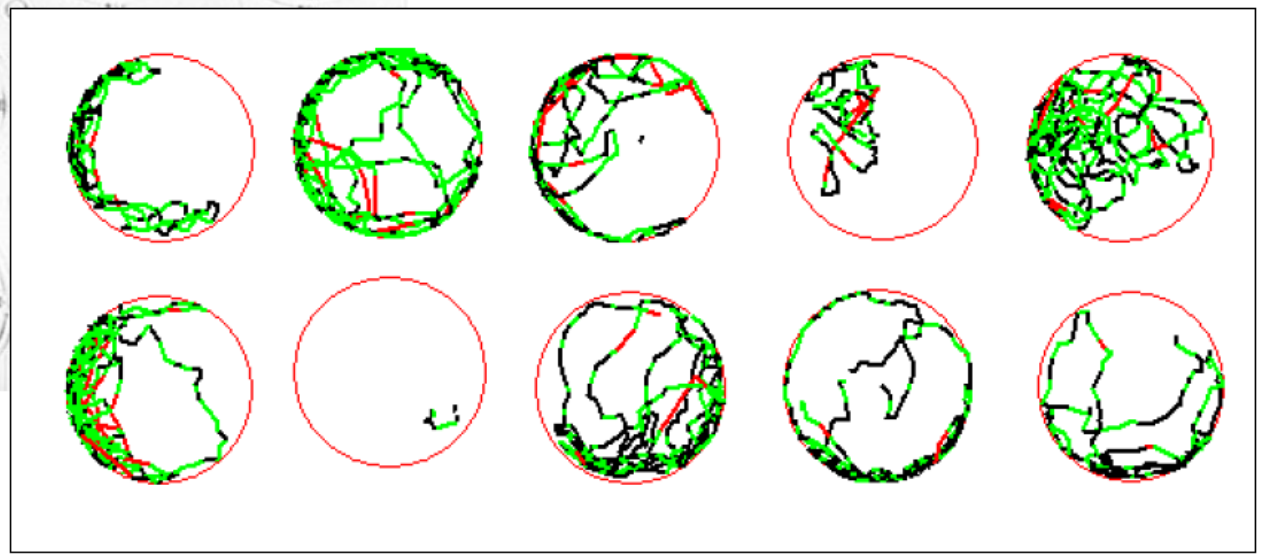
Zebrafish model:

- Physically underdeveloped
- Slow touch response
- Seizures
- Autistic features

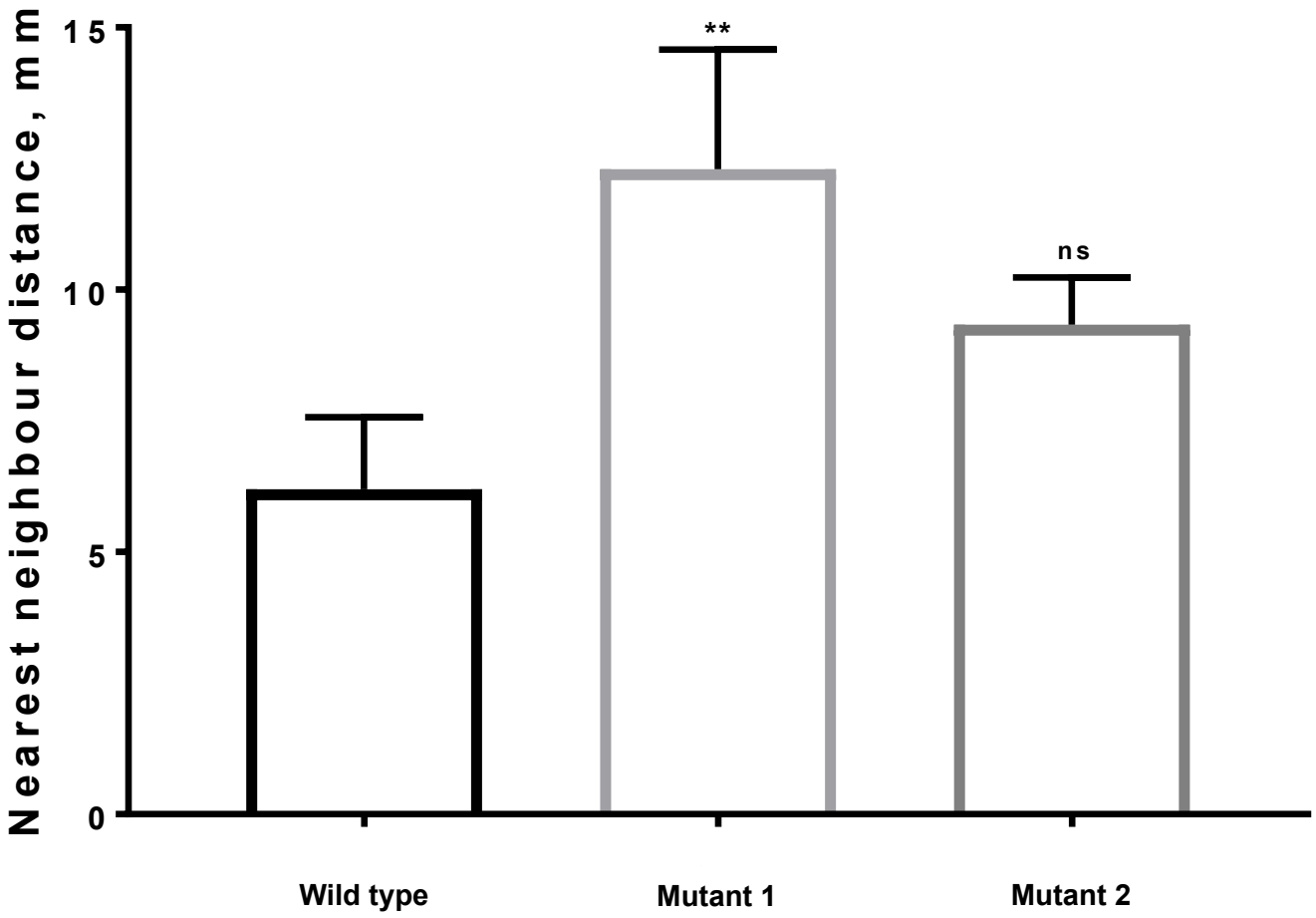
Behavioral larval locomotor assay (automated video tracking)



Movement Speed Example
Black: < 4mm/sec
Green: 4-20 mm/sec
Red: > 20 mm/sec



Altered shoaling behavior in genetic zebrafish mutant



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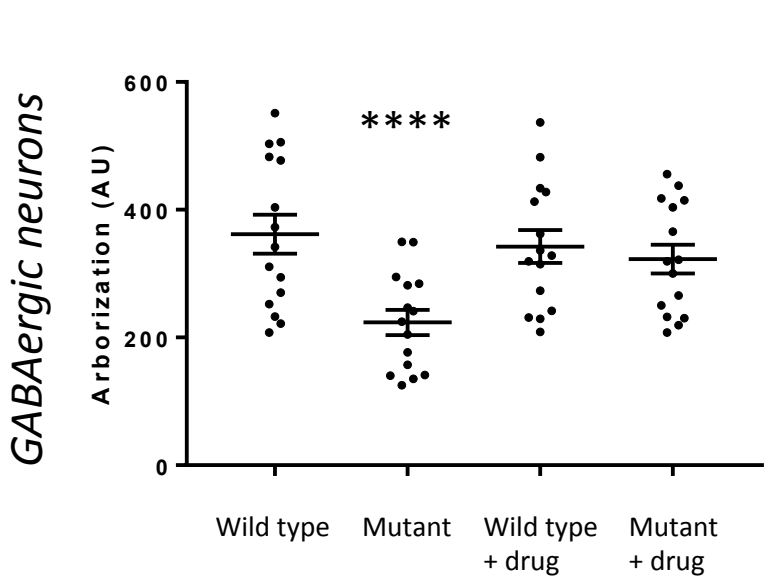
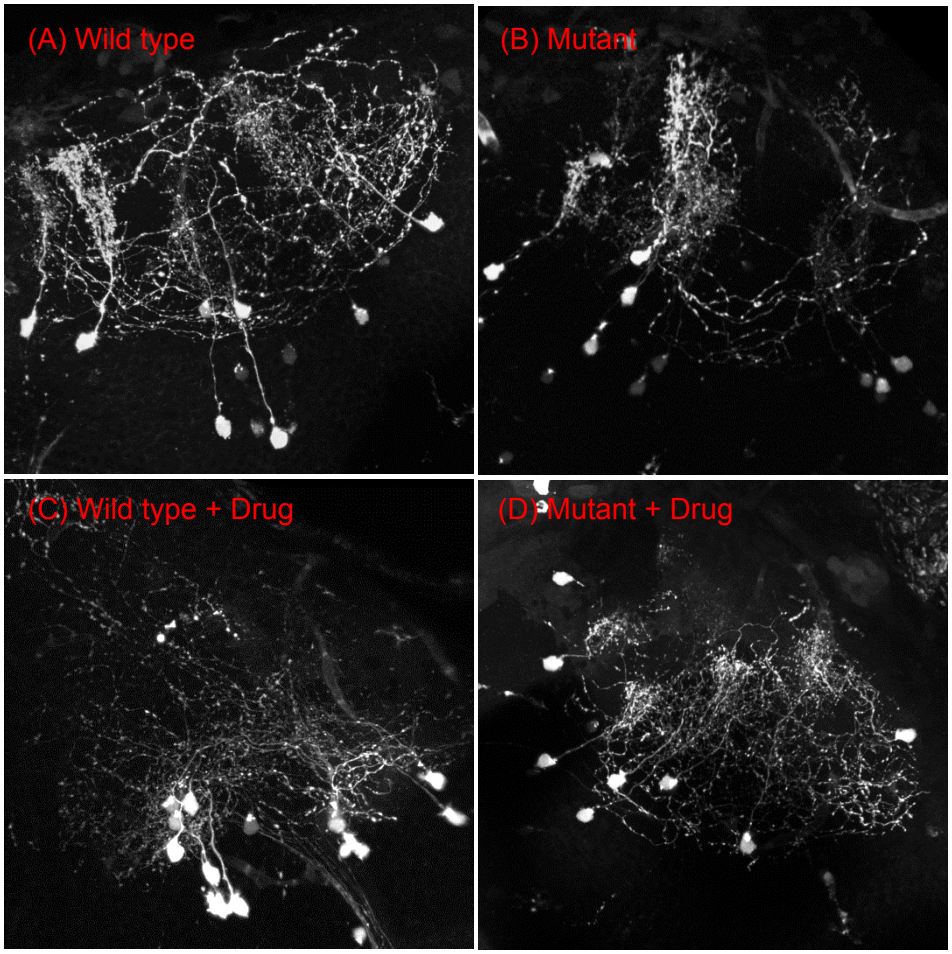
Neuronal branching defects in genetic mutant





Chronic drug treatment

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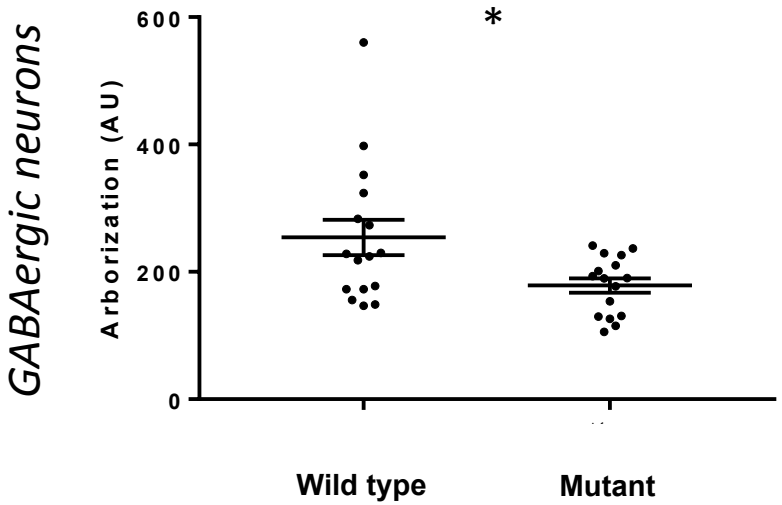
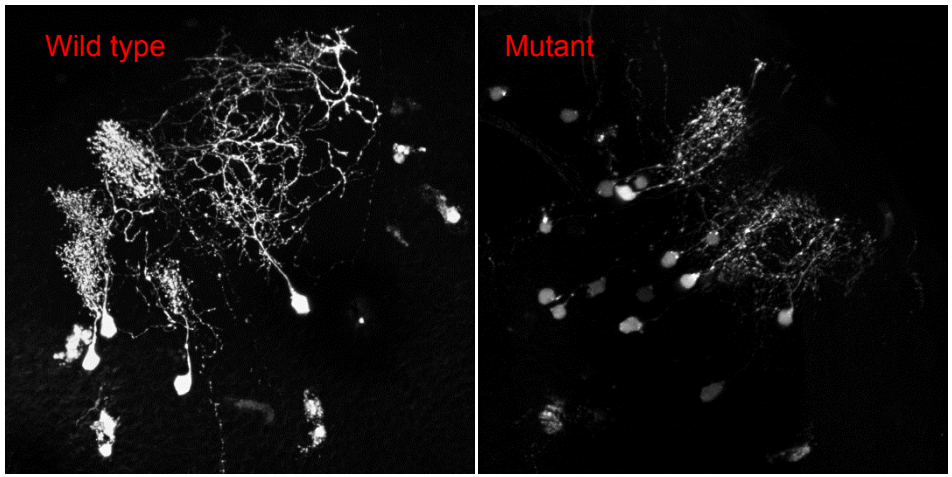
Neuronal structural defects precede seizures





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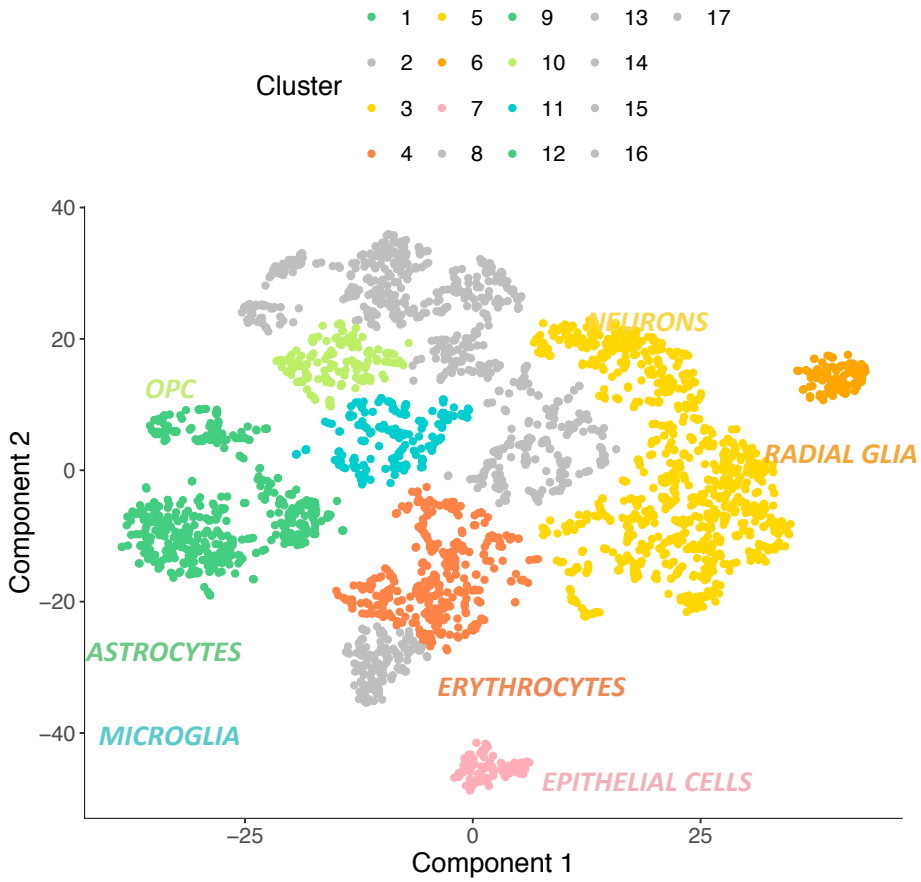
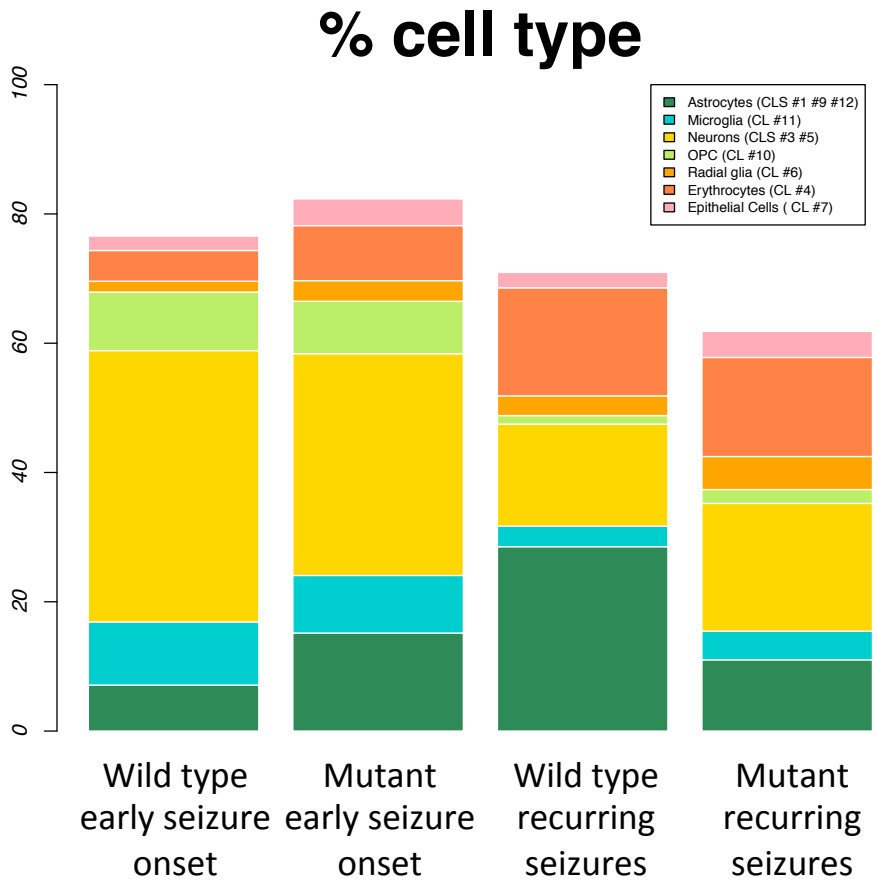
Gene expression and cell cluster profiling





In collaboration with A. Skupin, Luxembourg Center for Systems Biomedicine

C. Esguerra, SLC6A1 Round Table, AES 2018

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Epilepsy models

PLOS ONE

RESEARCH ARTICLE

Pharmacological Characterization of an Antisense Knockdown Zebrafish Model of Dravet Syndrome: Inhibition of Epileptic Seizures by the Serotonin Agonist Fenfluramine

Yifan Zhang[1☯], Angéla Kecskés[1☯], Daniëlle Copmans[1‡], Mélanie Langlois[2‡], Alexander D. Crawford[1,2], Berten Ceulemans[3], Lieven Lagae[4], Peter A. M. de Witte[1]*, Camila V. Esguerra[1¤]

Contents lists available at ScienceDirect

Epilepsy & Behavior

journal homepage: www.elsevier.com/locate/yebeh

Cross-species pharmacological characterization of the allylglycine seizure model in mice and larval zebrafish

Karine Leclercq [a,1], Tatiana Afrikanova [b,1], Melanie Langlois [c], An De Prins [d], Olivia E. Buenafe [b], Chiara C. Rospo [a], Ann Van Eeckhaut [c], Peter A.M. de Witte [b], Alexander D. Crawford [b,c], Ilse Smolders [d], Camila V. Esguerra [b,e,*], Rafal M. Kaminski [a,**]

AJHG

Cell PRESS

Volume 93, Issue 5, 7 November 2013, Pages 967–975

Report

De Novo Loss-of-Function Mutations in *CHD2* Cause a Fever-Sensitive Myoclonic Epileptic Encephalopathy Sharing Features with Dravet Syndrome

Arvid Suls[1, 2, 38], Johanna A. Jaehn[3, 38], Angela Kecskés[4, 38], Yvonne Weber[5, 38], Sarah Weckhuysen[1, 2], Dana C. Craiu[6, 7], Aleksandra Siekierska[4], Tania Djémié[1, 2], Tatiana Afrikanova[4], Padhraig Gormley[8], Sarah von Spiczak[3], Gerhard Kluger[9], Catrinel M. Iliescu[6, 7], Tiina Talvik[10, 11], Inga Talvik[10, 11], Cihan Meral[12], Hande S. Caglayan[13], Beatriz G. Giraldez[14], José Serratosa[14], Johannes R. Lemke[15], Dorota Hoffman-Zacharska[16], Elzbieta Szczepanik[17], Nina Barisic[18], Vladimir Komarek[19], Helle Hjalgrim[20, 21], Rikke S. Møller[20], Tarja Linnankivi[22],

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PLOS ONE

Validation of the Zebrafish Pentylenetetrazol Seizure Model: Locomotor versus Electrographic Responses to Antiepileptic Drugs

Tatiana Afrikanova[1☯], Ann-Sophie K. Serruys[1☯], Olivia E. M. Buenafe[1], Ralph Clinckers[2], Ilse Smolders[2], Peter A. M. de Witte[1], Alexander D. Crawford[1], Camila V. Esguerra[1]*

LETTERS

nature genetics

Mutations in *STX1B*, encoding a presynaptic protein, cause fever-associated epilepsy syndromes

Julian Schubert[1,30], Aleksandra Siekierska[2,30], Mélanie Langlois[3], Patrick May[3,4], Clément Huneau[5,6], Felicitas Becker[1], Hiltrud Muhle[7], Arvid Suls[8,9], Johannes R Lemke[10,11], Carolien G F de Kovel[12], Holger Thiele[13], Kathryn Konrad[13], Amit Kawalia[13], Mohammad R Toliat[13], Thomas Sander[13], Franz Rüschendorf[14], Almuth Caliebe[15], Inga Nagel[15], Bernard Kohl[16], Angela Kecskés[2], Maxime Jacmin[3], Katia Hardies[8,9], Sarah Weckhuysen[8,9], Erik Riesch[10,17,18], Thomas Dorn[19], Eva H Brilstra[12], Stephanie Baulac[20,21], Rikke S Møller[22,23], Helle Hjalgrim[22,23], Bobby P C Koeleman[12], EuroEPINOMICS RES Consortium[24], Karin Jurkat-Rott[25], Frank Lehmann-Horn[25], Jared C Roach[4], Gustavo Glusman[4], Leroy Hood[4], David J Galas[3,4,26], Benoit Martin[5,6], Peter A M de Witte[2], Saskia Biskup[17,18], Peter De Jonghe[8,9], Ingo Helbig[7], Rudi Balling[3], Peter Nürnberg[13,27,28], Alexander D Crawford[2,3], Camila V Esguerra[2,29,31], Yvonne G Weber[1,31] & Holger Lerche[1,31]

BRAIN

A JOURNAL OF NEUROLOGY

CHD2 variants are a risk factor for photosensitivity in epilepsy

Elizabeth C. Galizia,[1,2,*] Candace T. Myers,[3,*] Costin Leu,[1,2,*] Carolien G. F. de Kovel,[4] Tatiana Afrikanova,[5] Maria Lorena Cordero-Maldonado,[5] Teresa G. Martins,[5] Maxime Jacmin,[5] Suzanne Drury,[6] V. Krishna Chinthapalli,[1,2] Hiltrud Muhle,[7] Manuela Pendziwiat,[7] Thomas Sander,[8] Ann-Kathrin Ruppert,[8] Rikke S. Møller,[9,10] Holger Thiele,[8] Roland Krause,[5] Julian Schubert,[11] Anna-Elina Lehesjoki,[12,13] Peter Nürnberg,[8] Holger Lerche[11] for the EuroEPINOMICS CoGIE Consortium,[#] Aarno Palotie,[14,15,16] Antonietta Coppola,[1,2,17] Salvatore Striano,[17] Luigi Del Gaudio,[17] Christopher Boustred,[6] Amy L. Schneider,[18] Nicholas Lench,[6] Bosanka Jocic-Jakubi,[19,20] Athanasios Covanis,[21] Giuseppe Capovilla,[22] Pierangelo Veggiotti,[23,24] Marta Piccioli,[25] Pasquale Parisi,[26] Laura Cantonetti,[27] Lynette G. Sadleir,[28] Saul A. Mullen,[29] Samuel F. Berkovic,[18] Ulrich Stephani,[7] Ingo Helbig,[7] Alexander D. Crawford,[5] Camila V. Esguerra,[30,31] Dorothee G. A. Kasteleijn-Nolst Trenité,[4] Bobby P. C. Koeleman,[4,$] Heather C. Mefford,[3,$] Ingrid E. Scheffer[18,29,$] and Sanjay M. Sisodiya[1,2,$]

Antiseizure hits from medicinal plants

LAUREATES: AWARDS AND HONORS, SCS FALL MEETING 2011 CHIMIA 2012, 66, No. 4 229
doi:10.2533/chimia.2012.229 Chimia 66 (2012) 229–232 © Schweizerische Chemische Gesellschaft

Zebrafish Bioassay-guided Microfractionation for the Rapid *in vivo* Identification of Pharmacologically Active Natural Products

Soura Challal[§a], Nadine Bohni[a], Olivia E. Buenafe[bc], Camila V. Esguerra[b], Peter A. M. de Witte[b], Jean-Luc Wolfender[a], and Alexander D. Crawford[b]

[§]SCS-DSM Award for best poster presentation

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Integration of Microfractionation, qNMR and Zebrafish Screening for the *In Vivo* Bioassay-Guided Isolation and Quantitative Bioactivity Analysis of Natural Products

Nadine Bohni[1☯], María Lorena Cordero-Maldonado[2,3☯], Jan Maes[2], Dany Siverio-Mota[2], Laurence Marcourt[1], Sebastian Munck[4], Appolinary R. Kamuhabwa[5], Mainen J. Moshi[5], Camila V. Esguerra[2], Peter A. M. de Witte[2], Alexander D. Crawford[2*¶], Jean-Luc Wolfender[1¶]

Epilepsy & Behavior 24 (2012) 14–22
Contents lists available at SciVerse ScienceDirect
Epilepsy & Behavior
journal homepage: www.elsevier.com/locate/yebeh

Anticonvulsant activity of bisabolene sesquiterpenoids of *Curcuma longa* in zebrafish and mouse seizure models

Adriana Monserrath Orellana-Paucar [a,b], Ann-Sophie K. Serruys [a], Tatiana Afrikanova [a], Jan Maes [a], Wim De Borggraeve [c], Jo Alen [c], Fabián León-Tamariz [b], Isabel María Wilches-Arizábala [b], Alexander D. Crawford [a], Peter A.M. de Witte [a,*], Camila V. Esguerra [a]

ACS Chemical Neuroscience Research Article pubs.acs.org/chemneuro

Zebrafish Bioassay-Guided Microfractionation Identifies Anticonvulsant Steroid Glycosides from the Philippine Medicinal Plant *Solanum torvum*

Soura Challal,[†,#] Olivia E. M. Buenafe,[‡,#] Emerson F. Queiroz,[†] Snezana Maljevic,[§] Laurence Marcourt,[†] Merle Bock,[§] Werner Kloeti,[†] Fabian M. Dayrit,[∥] Alan L. Harvey,[⊥] Holger Lerche,[§] Camila V. Esguerra,[‡] Peter A. M. de Witte,[‡,#] Jean-Luc Wolfender,[*,†,#] and Alexander D. Crawford[‡,#]

ACS Chemical Neuroscience Research Article pubs.acs.org/chemneuro

Tanshinone IIA Exhibits Anticonvulsant Activity in Zebrafish and Mouse Seizure Models

Olivia Erin Buenafe,[†] Adriana Orellana-Paucar,[†,‡] Jan Maes,[†] Hao Huang,[§] Xuhui Ying,[¶,○] Wim De Borggraeve,[⊥] Alexander D. Crawford,[†,∥] Walter Luyten,[▽] Camila V. Esguerra,[*,†] and Peter de Witte[*,†]

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Insights from Zebrafish and Mouse Models on the Activity and Safety of Ar-Turmerone as a Potential Drug Candidate for the Treatment of Epilepsy

Adriana Monserrath Orellana-Paucar[1,2], Tatiana Afrikanova[1], Joice Thomas[3], Yelaman K. Aibuldinov[3], Wim Dehaen[3], Peter A. M. de Witte[1], Camila V. Esguerra[1*]

1 Laboratory for Molecular Biodiscovery, Department of Pharmaceutical and Pharmacological Sciences, University of Leuven, Leuven, Belgium , **2** Facultad de Ciencias Químicas, Escuela de Bioquímica y Farmacia, Universidad de Cuenca, Cuenca, Ecuador, **3** Laboratory for Molecular Design and Synthesis, Department of Chemistry, University of Leuven, Leuven, Belgium