INTERNATIONAL BEHAVIOURAL AND NEURAL GENETICS SOCIETY

6th Annual Meeting
June 9-12, 2005
Miramar Complex
Sitges, Spain

PROGRAM and ABSTRACTS

Thursday, June 9

14:00-16:00 On-site registration available

17:00-18:00 Guided tour and wine tasting in Torres wineries.
A bus to the Atlantida's reception will be available after the visit

19:00-22:00 Reception/Concert – Atlantida open-air discothèque on the sea

Friday, June 10

08:30-16:00 Registration available

08:30-08:45 Opening Remarks

08:45-10:00 **Plenary** by Dr. Susan Alberts, Duke University and the Institute for Primate Research at the National Museums of Kenya in Nairobi, ***Kin recognition in animals: insights about modes and mechanisms.***

10:00-10:30 Coffee Break

10:30-12:30 **Symposium I: Monitoring and modifying genes to understand memory and social behavior**; Chairs: Catharine Rankin and K. Peter Giese

Josh Dubnau - Cold Spring Harbor Laboratory, NY, USA - *Anatomical dissection of memory phases in Drosophila*

Mario de Bono - Medical Research Council Laboratory of Molecular Biology, Cambridge, UK - *Genes that influence social behavior in C. elegans*,

Catharine Rankin - University of British Columbia, Vancouver, BC - *Neurotransmitters, receptors and memory in C. elegans*

Karim Nader - McGill University, Montreal, Canada - *The labile nature of consolidated memories*

K. Peter Giese - University College London, UK - *Memory reconsolidation engages only a subset of immediate-early genes induced during consolidation*

Cristina Alberini - Mount Sinai School of Medicine, NY, USA - *Mechanisms underlying memory stabilization*

12:30-12:45 Symposium I discussion

12:45-14:00 Lunch

14:00-16:00 **Paper Session** (speaker is underlined and speaker's institution is listed)

Oliver Ambrée, N Görtz, K Keyvani, R Palme, W Paulus, C Touma, N Sachser - University of Münster, Germany - *Behavioural alterations precede amyloid pathology in TgCRND8 Alzheimer mice*

V Campuzano, Susana Pezzi, J Lucena, C Carreiro, E Aso, MC Valero, I Barthelemi, JL Barbero, LA Pérez-Jurado - Universidad Pompeu-Fabra, Barcelona, Spain - *Williams Syndrome: use of murine models as a tool to dissect cognitive and physical phenotypes*

Mary-Anne Enoch, I Belfer, LS Schwartz, H Hipp, B Albaugh, M Virkkunen, MB Max, D Goldman - National Institute on Alcohol Abuse and Alcoholism, NIH, USA - *Anxious temperament mediates linkage of GABRA2 and GAL haplotypes to alcoholism*

Cathy Fernandes, E Hoyle, E Dempster, LC Schalkwyk, D Collier - King's College London, UK -*Alpha 7 nicotinic receptor impairs working/episodic-like memory*

in the delayed matching-to-place (DMP) version of the Morris water maze in mice

F. Scott Hall, S Waters, I Sora, DL Murphy, K-P Lesch, GR Uhl - National Institute on Drug Abuse, NIH, USA - *Increased anxiety in serotonin transporter knockout mice: interaction with gene knockout of other transporters*

Christopher Janus, D Westaway - Mayo Clinic, Jacksonville, FL, USA - *Impaired associative learning in a mouse model of tauopathy*

Hee-Sup Shin - National CRI Centre for Calcium & Learning, KIST, Seoul, Korea - *Thalamic bursts and sensory gating in the mouse*

William Siesser, MP McDonald - Vanderbilt University, Nashville, TN, USA - *Hyperactivity and a reduced response to psychostimulants in the thyroid receptor ß PV knock-in mice*

16:00-18:00 Poster Session I (with refreshments): poster numbers 1-24

Saturday, June 11

08:30-17:00 Registration available

08:45-10:00 **Plenary** by Dr. Gerald McClearn, renowned geneticist of the Center for Developmental and Health Genetics at The Pennsylvania State University, ***The genetics of behavioural aging*.**

10:00-10:30 Coffee Break

10:30 -12:30 **Symposium II: Adult neurogenesis: genetics and function**; Chairs: Dan Goldowitz and Nancy L. Hayes

Nancy L. Hayes - UMDNJ-Robert Wood Johnson Medical School, NJ, USA - *Genetic dissection of stem cell proliferation and differentiation in the dentate gyrus*

Dan Goldowitz - University of Tennessee, Tennessee, USA - *Genetic architecture of the rostral migratory stream (RMS)*

Pierre-Marie Lledo - Pasteur Institute, Paris, France - *Nature and nurture in adult neurogenesis*

12:30-12:45 Symposium II Discussion

12:45-14:00 Lunch

14:00-16:00 **Symposium III: Alcohol withdrawal severity and drinking: are they genetically related?** Chairs; John Crabbe and Cindy Ehlers

John Crabbe - Portland VA Medical Center and Oregon Health & Science University, OR, USA - *Multivariate analyses of drug self-administration and withdrawal severity in inbred mouse strains*

Julia Chester - Purdue University, IN, USA - *Relationship between genetic differences in alcohol drinking and alcohol withdrawal in selectively bred rodent lines*

Christoph Fehr - University of Mainz, Mainz, Germany - *Principal component analysis of human alcohol withdrawal symptoms and their relationship towards alcohol dependence severity*

Cindy L Ehlers - The Scripps Research Institute, CA, USA - *Linkage analyses of drinking symptoms severity and alcohol withdrawal in Mission Indians*

16:00-16:15 Symposium III Discussion

16:15-17:15 IBANGS Business Meeting

17:15-19:15 Poster Session II (with refreshments): poster numbers 25-49

Sunday, June 12

08:30-16:00 Registration available

08:30-10:30 **Symposium IV: The genetic basis of phenotypic variation in behavior in *Drosophila melanogaster***; Chair, Robert R. H. Anholt

Robert R. H. Anholt - North Carolina State University, Raleigh, NC, USA - *The genetic architecture of odor-guided behavior in Drosophila*

Ralph J. Greenspan - The Neurosciences Institute, San Diego, CA, USA - *The fruits of behavioral selection in the fruit fly*

Charalambos P. Kyriacou - University of Leicester, UK - *Natural variation in clock genes of Drosophila*

Trudy F. C. Mackay - North Carolina State University, Raleigh, NC, USA - *Quantitative genetics of locomotor behavior in Drosophila*

10:30-10:45 Symposium IV Discussion

10:45-11:15 Coffee Break

11:15-13:15 **Invited Talks – *Outstanding Young Investigator Awardees***

Doo-Sup Choi - University of California, San Francisco, CA, USA - *Resistance to ethanol intoxication in mice lacking protein kinase C delta*

Michael Galsworthy - University of Zurich, Switzerland - *Identifying consistent and informative behaviours in mouse exploration tasks*

Helen Kamens – Oregon Health & Science University, Portland, OR, USA - *Mice selectively bred for high and low acute locomotor response to methamphetamine do not differ in ethanol-induced stimulation, sensitization or metabolism*

Lin Liu - King's College London, UK - *Serotonin N-acetyltransferase (Aanat) as a candidate gene for mouse baseline locomotor activity measured in the home cage*

13:15-13:30 Discussion

13:30-15:00 Lunch

15:00-17:00 **Symposium V: Deciphering the codes of cognition** Chairs, Mara Dierssen, Oliver Stork, Carmen Sandi

Javier de Felipe - Cajal Institute, CSIC, Madrid, Spain - *Reflections on the structure of the cortical minicolumn*

Lukas Pezawas - Genes, Cognition and Psychosis Program, NIMH, NIH, IRP, USA - *Genetic variation in BDNF function affects human hippocampal structure and function related to episodic memory*

Susan Sangha - University of Calgary, Alberta Canada - *Memory formation in a snail is dependent upon one specific cell*

Sabrina Davis - CNRS, Université Paris Sud, Paris, France - *MAP kinase signaling cascade and consolidation and reconsolidation of memory*

Oliver Stork - Inst of Physiology, Otto-von-Guericke University Magdeburg, Germany - *Molecular and genetic mechanisms of fear memory consolidation*

17:15-17:30 Symposium V Discussion

17:30-18:00 Closing Remarks

18:00-20:00 BREAK

20:00 Banquet – Posit Restaurant

Sponsored by

National Institute on Alcohol Abuse and Alcoholism (NIAAA),
National Institute of Mental Health (NIMH),
National Institute on Aging (NIA),
National Institute of Child Health and Human Development (NICHD)
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Plenary Speaker Abstracts

Plenary Session I

Kin recognition in animals: insights about modes and mechanisms.
SC Alberts
Most highly social animals show clear kin biases in behavior: they tend to bias mating behavior away from kin, presumably due to costs of inbreeding, and they tend to bias beneficent behavior towards kin, presumably because they accrue inclusive fitness benefits by doing so. Some mechanism of kin discrimination is required for animals to achieve this biasing. Traditionally, four mechanisms have been described, and have been considered to be quite distinct; two have been referred to as mechanisms involving learning, and two have been referred to as "genetic" mechanisms. Increasingly, however, researchers are recognizing that these four mechanisms represent points on a continuum, and the distinctions that have been drawn between them are misleading: all have a genetic component, and all have a learned component. Furthermore, each mechanism represents an *estimate* of kinship, and each has distinct sources of error. Using data from our work on wild primates and recent work from other labs on a range of mammalian species, I describe these sources of error, which are both genetic and behavioral. I develop the idea that the question "which mechanism do animals use to recognize kin" should be replaced with the question "how do animals integrate information from several sources of information to estimate kinship?"
Department of Biology, Duke University, Durham NC, USA and the Institute for Primate Research at the National Museums of Kenya in Nairobi.
Support for this research was provided by NSF IBN0322613 and NSF BCS0323553.

Plenary Session II

The genetics of behavioural aging.
GE McClearn
Most phenotypes of interest to behavioral sciences are continuously distributed outcomes of complex causal fields. These fields often feature multi-step diverging and converging pathways of influence arising from multiple inputs from both genetic and environmental sources, together with feed-back mechanisms. These features can generate intricate and powerful interactions among genetic factors, among environmental factors, and between these domains. Empirical data documenting such interactions in behavioral phenotypes have long been available, and new examples are appearing at an accelerating rate. These interactions illuminate the contextual dependence of genetic influences; the effect of an allelic substitution at a given locus may depend extremely and dramatically upon genotypes at other loci and upon the environmental milieu. Furthermore, the relative influences of genetic and environmental influences may change substantially with age. Examples of these effects will be presented and the implications that can be drawn with respect to the design and interpretation of functional genomics research in behavioral aging will be explored.
Center for Developmental and Health Genetics and Department of Biobehavioral Health, The Pennsylvania State University, University Park PA, USA.

Symposium Speaker Abstracts

Symposium I: Monitoring and modifying genes to understand experience dependent changes in behavior

Symposium abstract
Chairs:Catherine Rankin, University of British Columbia, Vancouver, BC, Canada; K. Peter Giese, University College London, UK
There are many different approaches used to study the ways that gene activation patterns are altered by experience. In the first part of this symposium we will present new techniques and approaches that have been developed in simple systems such as C. elegans and Drosophila that have led to new insights about different mechanisms of memory. Dr. de Bono will describe studies on social aggregation in C. elegans that have led to an investigation of how C. elegans detects and reacts to changing oxygen levels in the environment. These studies have demonstrated that C. elegans can remember and return to an oxygen level associated with food. Dr. Dubnau and Dr. Rankin will discuss how different mechanisms and/or different brain structures support different types of lasting memories for classical conditioning (Dubnau) and for habituation (Rankin). In the second part of this symposium we will present recent findings on a phenomenon called reconsolidation. Reconsolidation is a postulated process that occurs once a consolidated memory is reactivated and interference with reconsolidation can erase the memory. The presentations will focus on reconsolidation studies in rodents. Dr. Nader will introduce the phenomenon of reconsolidation. He will discuss the implications on memory storage and treatments of disorders resulting from traumatic memories. Dr. Giese will present an analysis of immediate-early gene expression to compare memory consolidation and reconsolidation at the molecular level. He will show that reconsolidation is a partial recapitulation of consolidation. Dr. Alberini will also present a molecular comparison of memory consolidation and reconsolidation and she will discuss a new working model that will explain recent controversies in the field.

Josh Dubnau, A.S. Chiang, Cold Spring Harbor Laboratory, USA
Title: Anatomical dissection of memory phases in Drosophila
One of the defining features of memory formation is the dynamic transfer of information between anatomical loci involved in short term versus long term storage. Even simple forms of Pavlovian learning in relatively simple invertebrate brains appear to involve information processing in a large network of neurons. We are using a genetic technique to map the circuitry involved in processing of Pavlovian associations in Drosophila. Using a genetically engineered variant of dynamin protein, we can transiently shut down specific groups of neurons in living animals. With this approach we can quickly take small groups of neurons in the brain offline and then moments later bring them online again. When combined with high resolution Confocal imaging, this technique allows us to investigate the circuitry involved in learning, in each phase of memory storage, and even in memory retrieval. With this method, we already have established that the circuitry responsible for Pavlovian memory in flies is substantially more complex than previously thought, and displays distinct neuronal circuitry for different phases of memory.

Mario de Bono, Medical Research Council Laboratory of Molecular Biology, Cambridge, England
Title: *C. elegans* responses to oxygen and other animals
Animal behaviour arises from the interaction between the environment, experience and the intrinsic properties of two dynamic networks - of genes and gene products and of neurons and neural circuits. We are studying how such networks encode C. elegans behavior and how they evolve. We can dissect neural networks into individual identified neurons since the C. elegans nervous system has exactly 302 neurons and these have reproducible functions and synaptic connections. C. elegans can come together to form groups. Aggregation is context- and experience-dependent. For example it can be elicited in response to food or following starvation. The decision to aggregate or disperse involves integration of multiple sensory cues, including signals from bacterial food, other animals, gases and internal nutritional state. Nociceptive neurons that detect harmful or repellent conditions stimulate aggregation. These neurons appear to act by counterbalancing the action of a different neural pathway that inhibits group formation. Neurons exposed to the body fluid of the animal regulate aggregation by integrating a different set of antagonistic cues. A pathway responding to food signals acts through a neuropeptide receptor to repress

aggregation, whereas a different pathway, mediated by soluble guanylate cyclases, measures oxygen levels to facilitate this behaviour. Neuroendocrine pathways whose activity is controlled by feeding status or a pheromone form additional layers of regulation. Aggregation behaviour is polymorphic in the wild providing an opportunity to investigate how behaviour evolves. Natural variation in C. elegans aggregation is associated with a single residue change in a neuropeptide G-protein coupled receptor. This change alters coupling between ligand and receptor and receptor and G-protein, and effectively reconfigures the sensory landscape of C. elegans. C. elegans displays surprising sophistication in its behavioural responses to food and other animals. We are studying the genetic and neural architecture that allows these responses to be integrated together.

Catherine H. Rankin and Jacqueline K. Rose, Department of Psychology and Brain Research Centre, University of British Columbia, Vancouver, BC, Canada

Title: *C. elegans* demonstrates a 12 hour memory that occurs independent of 24 hour long-term memory

C. elegans is capable of long-term memory (>24 hours) for habituation training to a mechanosensory stimulus; a tap to the side of the worm's petri plate. This memory requires a distributed (spaced) training protocol (80 taps delivered in 4 blocks of 20 taps) and is protein synthesis dependent. If tested earlier (i.e., 12 hours after training) we find a memory phase that is similar to Drosophila's anaesthesia-resistant memory (ARM). As reported in Drosophila, 12-hour retention in C. elegans can be produced by a massed (one training block of 80 taps) training protocol and does not require protein synthesis. Further we have found, unlike long-term memory, glutamate transmission does not appear to be required for 12-hour retention as mutant strains that effect glutamate transmission, i.e., eat-4 (glutamate vesicular transporter) and glr-1 (40% homology to the AMPA receptor), show no deficit in 12-hour retention. We have previously reported using confocal imaging of transgenic worms expressing GLR-1::GFP that worms that received distributed training had significantly less GLR-1::GFP expression 24 hours after training than control worms. When we examine GLR-1::GFP expression 12-hours after a massed training protocol, we see no difference in GLR-1::GFP expression compared to controls. We also looked for presynaptic changes using a transgene that expressed synaptobrevin-GFP in the tap sensory neurons and found an increase in synaptobrevin in trained worms compared to controls worms 12 hours after training, and no difference 24 hours after training. To determine what the additional vesicles contained 12 hours after massed training we investigated whether inhibitory neuropeptides expressed in the sensory neurons might contribute to 12 hour memory. Worms with mutations in genes for an FMRFamide gene did not express memory 12 hours after massed training. These data support the hypothesis that there are two separable forms of memory present at 12 and 24 hours post-training: one depends on protein synthesis and a decrease in glr-1 expression, the other does not require protein synthesis and appears to depend on an increase in the release of an inhibitory neuropeptide.

Karim Nader, McGill University, Canada

Title: The labile nature of consolidated memories

Memory consolidation theory posits that new memories initially enter a sensitive state during which they can be disrupted, a state called short-term memory (STM). Over time this STM is converted to a long-term memory (LTM) that is resistant to being disrupted. In order for memories to enter LTM, the neurons mediating the memory must produce new proteins that will be used for the long-term storage of the memory. Once information is in LTM, it is posited to be "fixed" in the brain. Recently, we showed that when a consolidated LTM is remembered or reactivated, it returns to a state similar to STM in that neurons must synthesize new proteins in order for the memory to persist. For example, if protein synthesis in neurons is inhibited after reactivation of a consolidated auditory fear memory, an old consolidated memory could be erased from the brain. This phenomenon is called Reconsolidation. The findings from my studies have significant clinical implications for memory disorders such as post-traumatic stress disorder (PTSD). Theoretically, reconsolidation challenges the foundation on which Memory Consolidation Theory rests. Current work in our lab is dedicated to understanding 1) whether there are boundary conditions for reconsolidation, 2) how the mechanisms mediating reconsolidation are similar or distinct from consolidation.

K. Peter Giese, University College London, UK

Title: Memory reconsolidation engages only a subset of immediate early genes induced during consolidation

The relationship between memory consolidation and reconsolidation at the molecular level is poorly understood. We performed a molecular analysis and identified three immediate-early genes that are differentially regulated in the mouse hippocampus after contextual fear conditioning and reactivation of the context-shock memory: Serum- and glucocorticoid-induced kinase 1 (SGK1), SGK3, and nerve growth factor-inducible gene B (NGFI-B). The upregulation of SGK1 expression was not specific for the context-shock association and therefore not suitable for a comparison of contextual memory consolidation and reconsolidation. SGK3 expression was upregulated during both consolidation and reconsolidation. Analysis of SGK3 expression showed that expression changes elicited by a context-shock association during consolidation can subsequently be recapitulated during reconsolidation and that the transcriptional changes induced by retrieval depend on the remoteness of the memory. On the other hand, we found that NGFI-B is regulated during consolidation but not reconsolidation. This consolidation-specific regulation occurs in hippocampal area CA1. Our discovery of a consolidation-specific transcription indicates that reconsolidation is only a partial recapitulation of consolidation at the transcriptional level. Such partial rather than total recapitulation may have evolved as a more economic and reliable mechanism for organisms to modify memory.

Critina Alberini, Mount Sinai School of Medicine, NY, USA

Title: Mechanisms of memory stabilization: are consolidation and reconsolidation similar or distinct processes?

The consolidation of new memories depends on a critical phase of protein synthesis. It is a widely held view that, once consolidated, memories are stable and resilient to disruption. However, this hypothesis has been challenged by the fact that established memories become labile when recalled and, to be maintained, require another phase of protein synthesis. Therefore, it has been proposed that each time a memory is reactivated it needs to undergo another process of consolidation (reconsolidation) in order to persist. To determine whether and to what extend reconsolidation is similar or different from the consolidation process, we have used molecular markers and investigated the temporal and anatomical requirements underlying both processes. I will discuss our recent results and a working model that may explain the apparently controversial findings of memory vulnerability after reactivation.

Symposium II: Adult Neurogenesis: Genetics and Function

Symposium abstract

Chairs: Dan Goldowitz, University of Tennessee, Memphis, Tennessee, USA; Nancy L. Hayes, UMDNJ-Robert Wood Johnson Medical School, Piscataway, NJ, USA

Neurogenesis in the adult mammal is confined to two regions of the brain: the subgranular zone of the dentate gyrus and the subventricular zone and rostral migratory stream adjoining the lateral ventricles. These two stem cell populations produce neurons that integrate into the circuitry of the adult brain over the lifespan of the animal. Disruption of adult neurogenesis is implicated in various human psychiatric disorders such as anxiety, depression and schizophrenia. In this symposium we present new genetic analyses of the development and proliferative behavior of these stem cell populations, analyses of the behavioral significance of these cells and their progeny, and their response to environmental and pharmacological influences.

Nancy L. Hayes, UMDNJ-Robert Wood Johnson Medical School, Piscataway, NJ, USA.

Title: Genetic Dissection of Stem Cell Proliferation and Differentiation in the Dentate Gyrus

Using inbred strains, we show that adult neurogenesis in the dentate gyrus is not a single phenomenon; rather, it is the net result of multiple phenotypes which are under separate genetic regulation. These include size and distribution of the stem cell population, proportion and distribution of their surviving progeny, and proportion of neurons produced. Analysis of recombinant inbred strains shows that QTL's for these traits can be identified and that there is little overlap in the regulation of proliferation in the dentate gyrus and in the rostral migratory stream.

Dan Goldowitz, University of Tennessee, Memphis, Tennessee, USA.

Title: Genetic architecture of the rostral migratory stream (RMS)
While many genes have been identified that can influence the proliferation and differentiation of adult progenitor cells in culture, the intrinsic genetic machinery of these cells is poorly understood. We have undertaken an analysis of recombinant inbred lines to localize regions of the genome that underlie the proliferative potential of the cells of the RMS. We find that the genetic hotspots for RMS proliferative capacity are different from those of the dentate gyrus, and some candidate loci are explored.

Luca Santarelli, Columbia University, New York, New York, USA
Title: Functional analysis of adult neurogenesis in the dentate gyrus
Psychiatric disorders are marked by anxiety and decreased cognition. In order to investigate the functional role of adult-born neurons, we altered adult hippocampal neurogenesis by genetic (transgenic, knockout) and radiological means and assessed the effects of these manipulations on hippocampal-dependent cognitive and emotional behaviors. We report that disruption of hippocampal neurogenesis causes cognitive and emotional deficits in behavioral tasks that require intact hippocampal function.

Pierre-Marie Lledo, Pasteur Institute, Paris, France.
Title: Nature and nurture in adult neurogenesis
The olfactory bulb represents the only brain area where local GABAergic neurons are continuously replaced. How the newborn neurons integrate into a pre-existing neural network and how basic functions are maintained when a large percentage of neurons are subjected to continuous renewal, are important questions that have attracted our attention. We shall see how the production of GABAergic interneurons is specifically adapted to experience-dependent regulation of neural networks and what are the programmed mechanisms molded by experience. In particular, we shall report the degree of sensitivity of the bulbar neurogenesis to the level of sensory inputs and, in turn, how the adult neurogenesis adjusts the neural network functioning to optimize sensory information processing. We will bring together recently described properties and emerging principles of interneuron function in the olfactory bulb that support a much more complex role for these cells than just providers of inhibition.

Symposium III: Alcohol Withdrawal Severity and Drinking: Are they Genetically Related?

Symposium abstract
Chairs: John C. Crabbe, Oregon Health & Science University, Portland, OR, USA; Cindy L. Ehlers, The Scripps Research Institute, La Jolla, CA, USA
Human alcohol withdrawal is a complex trait with heterogeneous symptoms such as hypertension, tachycardia, hyperhidrosis, agitation, anxiety, nausea, vomiting, auditory disturbances, visual disturbances, disturbances of orientation and withdrawal seizures. Symptom presentation and severity can largely differ between patients, but the biological basis is not well known yet. Data are emerging from a number of species that alcohol withdrawal is genetically regulated. However, its relationship to alcohol dependence severity, family history of alcoholism and severity of drinking is complex. This symposium will present current data on the genetics of alcohol drinking and withdrawal in three species (mouse, rat, human) using different genetics approaches including, inbred lines, candidate genes and linkage analyses. Emphasis will be placed on translational findings and common themes. All speakers have agreed to attend the meeting and present their talk.

John C. Crabbe, Portland Alcohol Research Center, Portland VA Medical Center and Oregon Health & Science University, Portland, Oregon, USA
Title: Multivariate analyses of drug self-administration and withdrawal severity in inbred mouse strains
Dr. Crabbe will describe data collected in 15 inbred mouse strains. Multivariate analyses show a strong negative genetic relationship between oral self-administration of diazepam solutions and the severity of diazepam withdrawal, precipitated by antagonist injection. A similar negative genetic relationship holds for ethanol, albeit weaker, but no such negative coupling could be seen for pentobarbital or morphine. On the other hand, withdrawal from all three drugs that affect the GABA-A receptor (i.e., excluding morphine) tended to reflect the influence of common genes. The data are consistent with findings from other genetic animal models and with human genetic epidemiological data suggesting that about half the

genetic risk for abuse of a given drug is unique to that drug, while the other half is generally shared among drugs.

Julia A. Chester, Department of Psychological Sciences, Purdue University, 703 Third Street, West Lafayette, IN, USA

Title: Relationship between genetic differences in alcohol drinking and alcohol withdrawal in selectively bred rodent lines

We examined the genetic relationship between alcohol drinking and sensitivity to alcohol withdrawal following acute alcohol treatment in both rat and mouse lines selectively bred for differences in voluntary alcohol intake. Male, low alcohol drinking rat lines (NP, LAD1, LAD2) demonstrated alcohol withdrawal signs but high alcohol drinking rat lines (P, HAD1, HAD2) did not using several different alcohol withdrawal indices. These results suggest that sensitivity to alcohol withdrawal is inversely related to a genetic propensity toward alcohol drinking, which agrees with findings in multiple genetic mouse models. Analysis of this genetic relationship was extended to mouse lines selectively bred for high (HAP) or low (LAP) alcohol preference using acoustic startle reactivity to index withdrawal from acute alcohol treatment. Male LAP but not HAP mice and female HAP but not LAP mice showed suppressed acoustic startle reactivity, suggesting reduced sensitivity to alcohol withdrawal. These findings are in contrast to prior results indicating greater alcohol withdrawal in LAP mice compared to HAP mice when handling-induced convulsions were used in index alcohol withdrawal. Overall, these data suggest that the genetic relationship between alcohol withdrawal and alcohol drinking may depend on sex, species, and the measure used to index alcohol withdrawal.

Christoph Fehr, Department of Psychiatry, University of Mainz, Untere Zahlbacher Str. 8, 55131 Mainz, Germany

Title: Principal component analysis of human alcohol withdrawal symptoms and their relationship towards alcohol dependence severity

Human alcohol withdrawal is a complex trait with heterogeneous symptoms of unknown relationship. Within the current project, we sought to identify common factors of human alcohol withdrawal. Seven different alcohol withdrawal symptoms (hypertension, tachycardia, psychomotor agitation, paroxysmal sweats, tremor, nausea, anxiety) were monitored with a modified German version of the CIWA-Ar alcohol withdrawal scale among 57 alcohol dependent inpatients over 8 days. In addition, the patients were asked for the occurrence and frequency of previous alcohol withdrawal seizures. Principal component analysis identified three main factors of human alcohol withdrawal. The first factor explained 38% of symptom variance and was predominately influenced by psychic symptoms of withdrawal such as psychomotor agitation, nausea and anxiety. Cardiovascular symptoms and sweating loaded on the second principal factor of alcohol withdrawal that explained 16% of the phenotypic variance. Alcohol withdrawal seizures loaded on a third independent factor. Correlation analyses of the withdrawal symptoms and different measures of dependence severity did reveal some positive (e.g. sum scores of the Michigan Alcohol Screening Test), negative (e.g., number of years of drinking) and nonsignificant findings. Taken together, human alcohol withdrawal seems to be influenced by at least three independent factors that show a very complex relationship towards other measures of alcohol dependence severity.

Cindy L. Ehlers, Department of Neuropharmacology, The Scripps Research Institute, 10550 N Torrey Pines Road La Jolla, CA, USA

Title: Linkage analyses of Drinking symptoms severity and alcohol withdrawal in Mission Indians

Alcohol dependence is a leading cause of morbidity and mortality in Native Americans, yet biological factors underlying the disorder in this ethnic group remain ellusive. Disorders like alcohol dependence may be influenced by a number of genes that may be difficult to detect because each has a small effect on the broad clinical phenotype. However, such genes might be detected if they have a major effect on a more narrowly defined phenotype. Aspects of alcohol-related behavior that have been long associated with severe alcoholism are withdrawal and tolerance phenomena, specified in DSM-IV (American Psychiatric Association 1994) as alcohol dependence with a "physiological component". In this study Linkage analyses were performed for alcohol withdrawal and drinking severity phenotypes. Alcohol use, dependence and withdrawal were made using the SSAGA. Variance component estimate methods were used to calculate LOD scores using SOLAR. Two chromosomes, 4 and 12, had peak LOD scores that

exceeded 2 for the alcohol use severity phenotype and three chromosomes 6, 15, 16 were found to have peaks with LOD scores that exceeded 2 for the withdrawal phenotype. No overlap was found between the two phenotypes. Evidence for linkage to chromosomes 4 and 15, and 16 have been reported previously for alcohol related phenotypes whereas no evidence has as yet been reported for chromosomes 6 and 12. The peak on chromosome 6 is in the general region of the GABBR1 receptor. These results corroborate the importance of several chromosomal regions highlighted in prior segregation studies in alcoholism and further identify new regions of the genome that may be unique to these phenotypes or this population of Mission Indians (supported by NIAAA10203, Center for health disparities and a grant from EGCRC/state of California).

Symposium IV: The genetic basis of phenotypic variation in behavior in *Drosophila melanogaster*

Symposium abstract
Chair: Robert R. H. Anholt, North Carolina State University, Raleigh, NC, USA
Behaviors are complex traits that arise from interactions between many segregating genes with effects that are sensitive to the environment. The genetic architecture of behavioral traits is composed of dynamic epistatic networks of pleiotropic genes that are influenced by interactions with the physical, social and sex environment. There is an increasing awareness that subtle changes in transcriptional regulation can elicit widespread "ripple" effects throughout the transcriptome with profound effects on behavior. In addition, naturally occurring allelic variants can evoke major changes in behavioral phenotypes. With Drosophila melanogaster one can rear large numbers of individuals under conditions of controlled genetic background and environment, allowing resolution of small effects on complex phenotypes at a level not readily attainable in other systems. This advantage together with its sequenced genome, its exquisite suitability to genetic manipulations, and a wealth of publicly available resources make Drosophila melanogaster an ideal model system for investigating the consequences of small alterations in gene expression on behavior. The proposed symposium is intended to exemplify the subtleties of hypomorphic mutational effects, of shifting allele frequencies during artificial selection, and of naturally occurring genetic variation on behavior using Drosophila as a model. The relationship between natural variation and adaptive evolution will also be addressed.

Robert R. H. Anholt, North Carolina State University, Raleigh, NC, USA
Title: The genetic architecture of odor-guided behavior in Drosophila
We used chemosensory behavior in Drosophila melanogaster, an essential survival trait, as a model to gain insights into the genetic architecture of behavior. Whole genome expression analysis of co-isogenic smell-impaired (smi) lines provided evidence for widespread transcriptional ripple effects throughout the genome as a consequence of the introduction of hypomorphic mutations. Analyses of transcriptional profiles showed that the genetic architecture of behavior is determined by dynamic and plastic epistatic networks of pleiotropic genes, some of which are sexually dimorphic in their expression, and that the behavioral phenotype manifests itself as an emergent property of such networks. Furthermore, transcriptional epistasis mirrored phenotypic epistasis. These observations raise the question of how evolutionary forces interact with such networks? To gain insight into this question we analyzed the multigene family of odorant binding proteins (OBPs), which are expressed in the antennal perilymph, where they are thought to interact with odorants. We asked whether different OBPs are subject to different evolutionary trajectories and whether polymorphisms in Obp genes are associated with phenotypic variation in chemosensory behavior. We sequenced two clusters of Obp genes, the Obp56 cluster and the Obp99 cluster, in 50 inbred lines from a natural population of Drosophila melanogaster as well as D. simulans and D. yakuba as outgroups. A total of 213 single nucleotide polymorphisms (SNPs) in 4 Obp99 genes and 229 SNPs in 9 Obp56 genes were identified. Population genetic analyses revealed significant departure from neutrality in some, but not all Obp sequences, suggesting that different OBPs have evolved along different evolutionary trajectories. Association analyses showed that - after correcting for the false positive discovery rate - 18 SNPs, confined to 3 genes of the Obp99 cluster (Obp99a,c and d), are significantly associated with variation in the avoidance response to the repellant odorant benzaldehyde.

Ralph J. Greenspan, The Neurosciences Institute, San Diego, CA, USA

Title: The fruits of behavioral selection in the fruit fly
Artificial selection in the laboratory is a classical method for detecting the presence of naturally occurring genetic variation in behavior. We have adapted the use of DNA microarrays to the analysis of the molecular phenotypes of Drosophila melanogaster strains behaviorally selected, in separate experiments, for differences in geotaxis and in aggression.

Charalambos P. Kyriacou, University of Leicester, United Kingdom
Title: Natural variation in clock genes of Drosophila
The circadian clock of Drosophila melanogaster is generated by a small number of dedicated clock genes which encode transcriptional regulators and kinases, as well as photoreceptors which entrain the clock to environmental light cycles. Subtle natural variation in a number of these genes reveals that, rather than being neutral, they have profound implications for Darwinian fitness. This results in a geographical differentiation of the relevant haplotypes due to the different types of selection that appear to be involved.

Trudy F. C. Mackay, North Carolina State University, Raleigh, NC, USA
Title: Quantitative genetics of locomotor behavior in Drosophila
Locomotion is an integral component of most animal behaviors, and impairments in locomotion accompany senescence and neurodegenerative disease. Locomotor activity is also a quantitative trait, exhibiting continuous phenotypic variation in populations that is attributable to multiple interacting quantitative trait loci (QTLs) with small individual effects that are sensitive to the environment. However, little is known of the genes and pathways affecting variation in locomotor behavior in natural populations. We developed a rapid and highly reproducible assay to quantify locomotor reactivity in response to an acute mechanical stress, and used this assay to map QTLs affecting variation in locomotor reactivity between a wild type line (Oregon) and a sluggish strain (2b) of Drosophila. We assessed locomotor reactivity for each of 98 recombinant inbred lines derived from these strains, and mapped four QTLs at cytological positions 1B;3E, 27B;29E, 30D;38A and 98A;99A by linkage to polymorphic roo transposable element insertion sites. As is typical for initial genome scans, the QTLs encompassed genomic intervals of 2600-9600kb, containing 300-1000 positional candidate genes. We used deficiency complementation mapping to fine-map the QTLs, and found the initial four QTLs fractionated into 12 smaller chromosomal regions containing on average 75 positional candidate genes. We identified 13 positional candidate genes affecting variation in locomotor reactivity between Oregon and 2b using complementation tests to mutations, including Dopa decarboxylase (Ddc) and Catecholamines-up (Catsup), which affect catecholamine biosynthesis. Finally, we used linkage disequilibrium mapping to show that common molecular polymorphisms in both these genes are associated with subtle, naturally occurring variation in locomotor reactivity.

Symposium V: Deciphering the codes of cognition

Symposium abstract
Chairs: Mara Dierssen, Genomic Regulation Center, Barcelona, Spain; Oliver Stork, Otto-von-Guericke University, Magdeburg, Germany; Carmen Sandi, EPFL, Lausanne, Switzerland
Cognitive functions are largely dependent upon both, genetic and developmental factors. This symposium presents recent developments in humans and animals aimed to understand the mechanisms that translate such genetic and environmental influences on brain structure and function. The development of novel genetic approaches and functional genomics technologies in the past years has inspired intensive work on mechanisms of learning and memory, and allowed to unravel the contribution of specific genes to various aspects of the mnemonic processing. Key advances on the topic will be reviewed in a variety of species, ranging from the snail (a single cell preparation to study the key proteins involved in memory storage), to rodents (rats, mice; including studies on gene expression changes and genetically modified animals) and humans (showing the drastic impact of a polymorphism in a single gene, in humans, on individual differences in encoding and retrieval).

Javier de Felipe, Cajal Institute, CSIC, Madrid, Spain

Title: Reflections on the structure of the cortical minicolumn

The anatomically defined minicolumn is a rather complex element of the cortical information processing circuit. It consists of assemblies of microcircuits established by discrete and regularly distributed vertical aggregates of pyramidal neurons. However, a given interneuron in a minicolumn does not only establish synapses with other cells of its minicolumn, but also with cells located in adjacent minicolumns. Furthermore, the pyramidal cell axon collaterals that make up the vertical aggregates by far surpass the size of the parent minicolumn. Thus, it seems clear that the general mechanisms of intracortical information processing are through the interactions of the minicolumns within a given macrocolumn and with other minicolumns belonging to distant macrocolumns. While many properties of the microcircuits within the minicolumn are stereotypical between cortical regions and species, others are unique to a given area or a set of areas within a given species, or even across orders. However, we have still to resolve which modifications are the result of evolutionary adaptations of excitatory and inhibitory circuits to particular functions? and how much information obtained by studying of non-human mammals can be applied to investigate the emergence of sophisticated cognitive functions in humans? Unfortunately, the gigantic task that is underway, of identifying all the elements of the macrocolumns in the functionally distinct areas of the human, is likely to be insufficient. This is in part due to the fact that some fundamental experimental data on the functional and anatomical organization cannot be obtained in humans for obvious ethical reasons. Thus, meaningful data must be obtained from experimental animals that can be used in computational models to analyze the functional complexity of circuits in the human neocortex. A starting point is to try to identify the basic microcircuits thought to be common to all species, and the variations specific to the human neocortex.

Lukas Pezawas, Genes, Cognition and Psychosis Program, NIMH, NIH, IRP, USA

Title: Genetic variation in BDNF function affects human hippocampal structure and function related to episodic memory

Introduction: BDNF plays critical roles in brain development and memory. A genetic variation in the BDNF gene (val66met) affects the function of BDNF in neurons and predicts variation in human memory. We hypothesized that consistent with the cellular and clinical effects of the BDNF val66met polymorphism and the role of BDNF in cortical development, met allele carriers would have reduced hippocampal gray matter volume. Methods: We investigated high-resolution anatomical magnetic resonance images (MRI) of 111 normal healthy volunteers (Caucasians of European ancestry) without any psychiatric life-time history using optimized voxel-based morphometry (VBM), a sophisticated fully automated morphological imaging technique, which allows for a statistical comparison of gray matter volume on a voxel-by-voxel basis. Results: Consistent with our initial hypothesis, we found bilateral reductions of hippocampal gray matter volume (right: $p<0.001$; left: $p=0.013$) in met-BDNF carriers compared with val/val-BDNF subjects. Furthermore, we performed an exploratory analysis of the entire brain and found that, compared to val/val-carriers, met-BDNF carriers exhibited additional loci of reduced gray matter volumes predominately in the lateral convexity of the frontal lobes, with peak values encompassing the dorsolateral prefrontal cortex bilaterally ($p<0.001$). Conclusion: The BDNF val/met polymorphism affects the anatomy of the hippocampus and the prefrontal cortex, and thus may be a modifying genetic factor in the expression of a number of normal and abnormal brain conditions dependent upon the development and plasticity of these critical brain systems.

Baquet et al. J Neurosci 24, 4250-8 (2004).
Egan et al. Cell 112, 257-69 (2003).
Gorski et al. J Neurosci 23, 6856-65 (2003).
Hariri et al. J Neurosci 23, 6690-4 (2003).

Susan Sangha, University of Calgary, Alberta Canada

Title: Memory formation in a snail is dependent upon one specific cell

It has been shown in several model systems that RNA and protein synthesis are necessary for long-term memory (LTM) consolidation. In addition to de novo protein and RNA synthesis, the soma of one specific cell is also required for LTM consolidation in the pond snail, Lymnaea stagnalis. Aerial respiration was operantly conditioned by applying a mildly aversive tactile stimulus to the respiratory orifice area whenever the snail attempted to perform aerial respiration thus leading to a decrease in this behaviour. A 3-neuron central pattern generator is necessary and sufficient for aerial respiratory behaviour. It is possible to ablate the soma of one of these neurons, RPeD1, leaving behind a functional and intact

neurite where local protein synthesis can occur. Successful RPeD1 soma ablation does not block the occurrence of aerial respiratory behaviour, as the remaining functional primary neurite is competent to mediate this behaviour. We show here that the soma of RPeD1 is necessary for LTM formation.
Once a memory has been consolidated into LTM it is resistant to disruption. However, if a memory is to be reactivated at a later time point it is thought to be transferred to an active and labile state. This memory is now vulnerable to disruption until it can be "reconsolidated" and transferred back to an inactive and resistant state. Here we show that reconsolidation also requires the presence of RPeD1 as well as protein and RNA synthesis. Does the memory still require these steps when it has been reactivated several times? Is it possible that a reactivated memory becomes resistant to disruption? We demonstrate that upon several memory reactivations a memory first becomes RNA synthesis and RPeD1 independent and then protein synthesis independent. These results may help explain why well-rehearsed memories are harder to forget than newer less-rehearsed memories.
This work was supported by the Natural Sciences and Engineering Research Council of Canada and the Canadian Institutes of Health Research.

Sabrina Davis, CNRS, Université Paris Sud, Paris, France
Title: MAP kinase signaling cascade and consolidation and reconsolidation of memory
Activation of the ERK/MAP kinase signalling cascade leads to the translocation of ERK from the cytosol to the nucleus where, via the transcription factors Elk-1 and CREB it activates immediate early genes(IEGs), such as zif268. It is well accepted that IEG's transcriptional activation is an early mechanism for triggering late genetic events that underlie the stabilisation of long-term synaptic plasticity and memory consolidation. Studies using mutant mice or pharmacological inhibition of ERK, CREB and zif268 show that disruption of this part of the pathway that links phosphorylation of ERK with triggering the activation of zif268, results in decremental synaptic plasticity and deficits in the long-term consolidation of memory. More recently with the rejuvenation of the concept of memory reconsolidation after retrieval, evidence is beginning to emerge to suggest this pathway may also be activated during the reactivation of already consolidated memories and is required for their reconsolidation. In this brief presentation, I will review the evidence supporting the necessary role of this part of the ERK/MAP kinase pathway in consolidation and reconsolidation of memories.

Oliver Stork, Institute of Physiology, Otto-von-Guericke University Magdeburg, Germany
Title: Molecular mechanisms of fear memory consolidation
The amygdala mediates effects of emotional relevance on information storage in other brain areas and itself comprises a site of neural plasticity during fear memory formation. We investigated the molecular mechanisms of fear memory formation using Pavlovian fear conditioning, a leading aversive learning paradigm that is critically dependent on the structural and functional integrity of the amygdala. Key molecules were identified of structural reorganisation and changes in neuronal connectivity in this brain area, including cell adhesion molecules, cytoskeleton elements and members of the ubiquitin / proteasome pathway. Expression of the novel serine/threonine kinase Ndr2 was specifically increased in the amygdala during consolidation of fear memory. Molecular and cellular characteristics of Ndr2 were determined in vitro and in cell culture and suggest a role as a modulator of actin filament dynamics in cell adhesion and neurite outgrowth. Ndr2 likely mediates Ca++ signals towards the actin cytoskeleton during spinogenesis and synapse formation, thereby closely interacting with small GTPases of the Rho family. Our studies demonstrate mechanisms related to a structural reorganisation of amygdala neurons, which should contribute to the increased neural activity and synchronisation in the amygdalo-hippocampal system of fear conditioned mice.
Supported by the Deutsche Forschungsgemeinschaft (SFB426) and the Academy and Science Programme of the State Sachsonia-Anhalt.

Paper Session Abstracts

Behavioural alterations precede amyloid pathology in TgCRND8 Alzheimer mice.
O Ambrée1, N Görtz1, K Keyvani2, R Palme3, W Paulus2, C Touma1, N Sachser1
Besides cognitive decline, non-cognitive behavioural symptoms are a major burden for people suffering from Alzheimer's disease (AD), as well as for their relatives and care-givers. The symptoms such as restlessness, sleep disturbances, wandering and agitation are categorised in the "Behavioural and Psychological Symptoms of Dementia" (BPSD), and are in fact the primary reason for institutionalisation of Alzheimer patients. Studies using animal models of AD only rarely focus on behavioural aspects other than learning tasks and other standard tests in unfamiliar environments. For that reason, the goal of this study was to investigate the spontaneous home cage behaviour of transgenic and wildtype animals of an APP-transgenic murine model of AD (TgCRND8) in order to look for possible analogies to BPSD. At 30, 60, 90 and 120 days of age, behaviour was analysed with respect to 24h activity patterns and stereotypic behaviour. Simultaneously, corticosterone metabolites (CM) were monitored from fecal samples. Transgenic and wildtype animals differed significantly with respect to activity patterns at all test days. Additionally, transgenic mice also clearly displayed more stereotypic behaviour than wildtypes, which correlated significantly at 90 and 120 days of age with elevated CM concentrations. Activity patterns in TgCRND8 mice resemble the altered rhythms of activity in AD patients. Stereotypic behaviours and non-cognitive behavioural symptoms of human AD may share the same neural mechanisms. It is likely that analogies to BPSD are found in APP-overexpressing TgCRND8 mice. Remarkably, these behavioural alterations are observed before amyloid pathology in this model usually appears (around 90 days of age).
1Department of Behavioural Biology, University of Münster, Germany. 2Institute of Neuropathology, University Hospital Münster, Germany. 3Institute of Biochemistry, University of Veterinary Medicine Vienna, Austria.

Williams Syndrome: use of murine models as a tool to dissect cognitive and physical phenotypes.
V Campuzano, S Pezzi, J Lucena, C Carreiro, E Aso, MC Valero, I Barthelemi, JL Barbero, LA Pérez-Jurado
Williams-Beuren syndrome (WBS) is a developmental disorder occurring in ~1/20000 live births, caused by the heterozygous deletion of ~1.5Mb on chromosome 7q11.23. The commonly deleted region encompasses 26-28 genes flanked by segmental duplications that predispose to the mutational mechanism in humans. The only phenotype unambiguously associated with deletion of a gene is supravalvular aortic stenosis and the elastin gene. However, detailed deletion mapping on atypical patients with smaller deletion has identified two genes (GTF2IRD1 and GTF2I), encoding members of a novel family of transcription factors, that are strong candidates for the main aspects of the cognitive phenotype. To investigate the putative role of the general transcription factor gene GTF2I, in the development of the cognitive phenotype we have used gene targeting in mice. In our model, the deletion of exon 2 of Gtf2i containing the start codon, abrogates to the translation of a modified TFII-I protein lacking the first 90 aminoacids. Previous in vitro studies have demonstrated that this mutated form is unable to bind DNA and does not activate transcription. Mutant mice are viable and display some craniofacial anomalies and mild postnatal growth retardation, obvious in the homozygous state. We are currently characterizing the neurocognitive and physical phenotype of these mutant mice.
Dpto. Ciencias Experimentales y de la Salud. Universidad Pompeu-Fabra. Barcelona. Spain.

Anxious temperament mediates linkage of GABRA2 and GAL haplotypes to alcoholism.
M-A Enoch, I Belfer, LS Schwartz, H Hipp, B Albaugh, M Virkkunen, MB Max, D Goldman
GABAA receptor activation mediates many behavioral effects of ethanol and benzodiazepines. The GABAAa2 gene (GABRA2) modulates anxiety and response to stress and has recently been linked with alcoholism. Animal studies have implicated the neuropeptide galanin in alcohol abuse and anxiety. The aim of our study was to investigate GABRA2 and GAL haplotype-based associations with alcoholism and anxious temperament in two diverse population isolates. DSM-III-R lifetime diagnoses were determined in 331 Plains American Indian men and women and 514 Finnish Caucasian men. TPQ harm avoidance (HA), a dimensional measure of anxiety, was obtained. Haplotype-based analyses of 9 GABRA2 SNPs identified

2 haplotype blocks. Two complementary haplotypes in block 2 accounted for 89-92% of haplotype diversity. Genotyping of 5 GAL SNPs identified a single haplotype block with 5 common haplotypes. Alcoholics had higher HA than non-alcoholics ($P < 0.005$). In both populations of men only, the more abundant GABRA2 block 2 haplotype was associated with high HA alcoholics (> or = mean HA of population), the less abundant haplotype was associated with low HA alcoholics (< mean HA of population), and non-alcoholics were intermediate ($p < 0.05$). The Finnish men showed GAL haplotype ($p = 0.001$) and diplotype ($p = 0.002$) associations with alcoholism. Distribution of the 5 common GAL haplotypes ($p < 0.0001$) and diplotypes ($p < 0.0001$) differed between high HA alcoholics, low HA alcoholics and non-alcoholics. Similar results were observed in the Plains Indian men but not in the women. Anxiety may mediate linkage of GABRA2 and GAL haplotypes with alcoholism in men. Male alcoholics with high and low dimensional anxiety may be genetically distinct subtypes.
Laboratory of Neurogenetics, NIAAA, NIH, Bethesda, MD 20892.

Alpha 7 nicotinic receptor impairs working/episodic-like memory in the delayed matching-to-place (DMP) version of the Morris water maze in mice.
C Fernandes1, E Hoyle1, E Dempster1,2, LC Schalkwyk1, D Collier1,2
Patients with schizophrenia exhibit deficits in a range of cognitive functions, particularly working and episodic memory, which are thought to be core features of the disorder. Memory dysfunction in schizophrenia is familial and a promising endophenotype for genetic studies. Both human and animal work suggest a role for the neural nicotinic receptor in cognition, and studies implicate the alpha-7 subunit of the nicotinic receptor in schizophrenia. Consequently, we tested mice lacking the α7 nicotinic receptor (B6.129S7-*Chrna7*^{tm1Bay}/J) in the delayed matching to place task (DMP) of the Morris water maze, a measure of working /episodic memory akin to human episodic memory. A significant impairment in the α7 knockout mice was observed in DMP task (GENOTYPE X TRIAL interaction $F[2,26]=2.2$, $p=0.05$). This suggests a role for the alpha-7 subunit in working/episodic memory and supports a role for the α7 neural nicotinic acid receptor, CHRNA7, in schizophrenia and its endophenotypes.
1) Social, Genetic and Developmental Psychiatry Centre and 2) Division of Psychological Medicine. Institute of Psychiatry, King's College London, UK.

Increased anxiety in serotonin transporter knockout mice: interaction with gene knockout of other transporters.
FS Hall1, S Waters1, I Sora2, DL Murphy3, K-P Lesch4, GR Uhl1
Gene knockout of the serotonin transporter (SERT) in mice increases behavioral measures of anxiety. These experiments were conducted to determine whether further gene deletions of the dopamine transporter (DAT) or the vesicular monoamine transporter 2 (VMAT2) would complement these changes that in SERT KO mice. Anxiety was assessed in double knockout mice produced by interbreeding of the single knockout strains using 2 tests of anxiety: an emergence test and a novelty test. Gene deletion of the DAT or VMAT2 alone had no effect on anxiety in any of these tests. Consistent with previous results deletion of SERT produced elevations in behavior indicative of anxiety in both tests. Combined DAT KO/SERT deletion, even a heterozygous DAT deletion, counteracted some effects produced by SERT deletion alone. Combined VMAT2 KO/SERT KO produced a similar, but not significant, trend, perhaps because the effect of the SERT KO alone was less pronounced in these mice. These data support the role of multiple genes in determining baseline anxiety, including both dopaminergic and serotonergic genes. Furthermore, they demonstrate the viability of this approach to examine gene-gene interactions important for the study of the polygenic nature of anxiety disorders and related co-morbid conditions.
1National Institute on Drug Abuse/NIH/DHHS USA, 2Tohoku University Japan, 3National Institute on Mental Health/NIH/DHHS USA, 4University of Wuerzburg Germany.

Impaired associative learning in a mouse model of tauopathy.
C Janus, D Westaway
Transgenic mice expressing an FTPD-17 mutation P301L of tau were generated using a position-independent prion cosmid. The TgTau(P301L)23027 mice recapitulate key features of the human pathology including neurofibrillary tangles (NFTs) in the frontal areas of the cerebrum, in the brainstem, and to lesser extent in the spinal cord. These features were accompanied by gliosis, granulovacuolar changes, spongiosis, neuronal loss and cerebral atrophy. Mutant forms of tau accumulating in the CNS of these Tg animals were found to be hyperphosphorylated, conformational altered, ubiquitinated and

sarkosyl-insoluble, with electron microscopy revealing twisted ribbon filaments. The TgTau(P301L)23027 did not show any physical developmental abnormalities nor deficiencies in reproductive behaviour. Their locomotor and exploratory activity were not compromised. Cognitive characterization of aged TgTau(P301L)23027 revealed no significant deterioration in their spatial reference learning and memory, including reversal of spatial learning, as evaluated in the Morris water maze. Interestingly, old TgTau(P301L)23027 showed a significant impairment in a cued (visible platform) version of the water maze. Moreover, these Tg mice were also impaired in the acquisition of taste aversion and showed a significantly accelerated extinction of taste aversion. Though expression of human tau was invariant between Tg animals, the levels of insoluble tau protein in the brain was highly variable and positively correlated with the impairment in the taste aversion and with the inferior performance in the cued water maze test. This remarkable deficiency in associative learning of the Tg mice may indicate either an extreme sensitivity to genetic background effects or the intervention of cryptic environmental variables, and offers a remarkable parallel to the phenotypic diversity of FTPD-17 itself. In conclusion, the association between tau pathology and cognitive behaviour in TgTau(P301L)23037 mouse model may present a unique opportunity to better understand the cascade of tau pathology and the role of modifiers in FTPD-17.
Mayo Clinic Jacksonville, USA and University of Toronto, Toronto, Canada.

Thalamic bursts and sensory gating in the mouse.
H-S Shin
The knock-out mouse deficient in the alpha1G T-type channel, a dominant form of low-threshold calcium channels in the thalamocortical neurons, lacks low threshold calcium currents and thus low threshold burst firing in the thalamocortical neurons. This mouse presents a unique opportunity to examine the physiological role of thalamic burst firings in vivo. The mutant mouse reveals phenotypes that can be explained by impairment in thalamic sensory gating. The mouse is resistant to both drug-induced and spontaneous absence seizures (1, 2) which are characterized by a transient block of sensory relay by the thalamus. The mutant shows enhanced nociceptive behaviors toward chronic pains as a result of persistently increased pain signals passing through the thalamus to the cortex (3). The mouse shows sleep disturbances with abundant occurrence of brief awakenings, reminiscent of parasomnia in humans (4). These three different phenotypes reflect impaired sensory gating due to the lack of burst firing in the mutant thalamus. In addition, other behavioral aspect of the mutant mice that is similar to affective disorders in humans will be discussed in the context of impaired sensory gating. References: (1) Kim, D. et al. (2001) Neuron 31: 35-45; (2) Song, I. et al. (2004) J. Neuroscience, 24(22): 5249-5257; (3) Kim, D. et al. (2003) Science, 302:117-119; (4) Lee, J. et al. (2004) PNAS, USA, 101(52):18195-18199.
National CRI Centre for Calcium & Learning, KIST, Seoul, Korea.

Hyperactivity and a reduced response to psychostimulants in the thyroid receptor ß PV knock-in mice.
WB Siesser2, MP McDonald1,2,3
The thyroid receptor ß PV knock-in (TRßPV KI) mice express a mutant TRß allele derived from a patient with the rare genetic disorder Resistance to Thyroid Hormone (RTH). Among other features, the majority of RTH patients meet the diagnostic criteria for Attention Deficit-Hyperactivity Disorder (ADHD). We have shown that the TRßPV KI mice are hyperactive in the open field after multiple sessions in the apparatus. We report herein that the TRßPV KI mice increase their locomotor activity during the course of a single session as opposed to the habituation observed in wild-type animals. Psychostimulants such as methylphenidate, amphetamine, and the dopamine-transporter-selective inhibitor GBR12909, which increase locomotor activity in wild-type littermates, had no effect or a reduced effect on the TRßPV KI mice. In addition, monoamine concentrations and transporter function were measured in the striatum, nucleus accumbens, and prefrontal cortex of the TRßPV KI mice. The finding of an altered response to psychostimulants in the TRßPV KI mice suggests changes in catecholamine function stemming from the mutant TRß allele. These changes in catecholamine function may be analogous to those that occur in children with ADHD.
1Department of Pharmacology, 2Program in Neuroscience, and 3John F. Kennedy Center for Research on Human Development, Vanderbilt University, Nashville, Tennessee, USA. This work was supported by NINDS (1R21NS043581-01A1), NICHD (5P30HD015052-23), and the Nicholas Hobbs Society.

Outstanding Young Investigator Abstracts

Outstanding Young Investigator – Junior Faculty
Resistance to ethanol intoxication in mice lacking protein kinase C delta.
D-S Choi, JK Deitchman, W-H Chou, VN Kharazia, RO Messing
In vitro, chronic ethanol exposure increases the abundance of protein kinase C delta (PKCd) and is involved in up-regulation of L-type calcium channels. We generated PKCd-null mice to investigate the role PKCd in responses to ethanol in vivo. PKCd-null mice displayed much less ethanol-induced rotarod ataxia, and decreased hypothermic and hypnotic responses to ethanol compared with wild-type littermates. In contrast, ethanol consumption and preference were similar in both genotypes. Ethanol-induced motor incoordination is thought to involve ethanol actions at GABAA receptors, NMDA receptors and adenosine A1 receptors. Using drugs that act selectively at these three receptors, we found that PKCd-null mice are resistant only to some but not all GABAA receptor modulators. PKCd-null mice showed reduced rotarod ataxia when treated with pentobarbital, neurosteroids, or THIP when compared to wild-type mice. In contrast responses to the benzodiazepine flunitrazepam were similar in will type and PKCd-null mice. This pharmacology suggests that PKCd modulates d subunit-containing extrasynaptic GABAA receptors in motor control circuits.
Ernest Gallo Clinic and Research Center, Department of Neurology, University of California, San Francisco, Emeryville, California, 94608. This work was supported by funds provided by the State of California for medical research on alcohol and substance abuse through UC San Francisco and by AA013588 to R.O.M.

Outstanding Young Investigator – Postdoctoral Fellow
Identifying consistent and informative behaviours in mouse exploration tasks.
MJ Galsworthy, R Madani, H-P Lipp, DP Wolfer
Over the past 10 years, more than 4,000 transgenic and wildtype mice of various genetic backgrounds have been run in up to 5 exploration tasks in our lab (open field, light/dark box, null maze, emergence, object exploration). Procedures remained standard throughout and the complete data were meticulously kept in a huge database. In this meta-analysis, we employ the full database in combination with factor analysis, cross-arena reliability indices, and time-course analysis. This allows identification of which behaviours and time-points reveal maximal information content in terms of cross-arena traits or sensitivity to individual and group differences. We also demonstrate a factor-analytic exploration of cross-arena traits free from confounding within-arena instrument covariance. Reliability analysis of >30 measures demonstrates substantial range in the consistencies of behaviour measures: Cumulative circling shows the highest inter-arena correlations at r=0.78, major exploration variables appear in the middle ranks, and at the low end are several ?ethological? micro-behaviors; scanning movements (r=0.12), vertical movements (r=0.05) and head dips/pokes (r=0.03). Analysis of between-group variance as a proportion of total variance also indicates when and within what measures the greatest genetic sensitivity lies. Conclusions centre on the use of psychometrics and quantitative genetics in discerning optimal behaviour measures and refining tasks.
Division of Neuroanatomy and Behavior, Department of Anatomy, University of Zurich, Switzerland. Supported by Swiss National Science Foundation and NCCR "Neural Plasticity and Repair".

Outstanding Young Investigator – Graduate Student
Mice selectively bred for high and low acute locomotor response to methamphetamine do not differ in ethanol-induced stimulation, sensitization or metabolism.
HM Kamens1,2, S Burkhart-Kasch1,2, CS McKinnon1,2, N Li1,2, C Reed1,2, TJ Phillips1,2,3
Mice were selectively bred for high (HMACT) and low (LMACT) sensitivity to an acute injection of methamphetamine. The HMACT and LMACT line differed significantly in their acute locomotor response to methamphetamine in the first selected generation and continued to differ over the four generations of selection. These lines were used for gene mapping and to identify traits that share genetic influence with the selection trait. Some evidence suggests that there is a genetic correlation between the acute locomotor response to ethanol and methamphetamine. Therefore, we tested if the HMACT and LMACT lines differed in their acute locomotor response to ethanol as well as ethanol-induced sensitization and ethanol metabolism. The HMACT and LMACT lines did not differ in sensitivity to ethanol-induced stimulation or in rate of ethanol clearance. Both the HMACT and LMACT lines show locomotor

sensitization to a 2 g/kg dose of ethanol, but the lines do not differ in magnitude of sensitization. These results suggest that common genes and neurochemical mechanisms do not underlie the acute locomotor response to methamphetamine and ethanol-induced locomotor stimulation, sensitization, or metabolism of ethanol.
1. Department of Behavioral Neuroscience, 2. Portland Alcohol Research Center, 3. VA Medical Center Research Service, Oregon Health & Science University, Portland, OR USA. Supported by: The Department of Veterans Affairs, NIDA RO1 DA10913, NIAAA P50 AA10760, and NIAAA Training Grant T32 AA07468.

Outstanding Young Investigator – Graduate Student
Serotonin N-acetyltransferase (Aanat) as a candidate gene for mouse baseline locomotor activity measured in the home cage.
L Liu, MJ Parsons, C Fernandes, JL Paya-Cano, LC Schalkwyk
As a quantitative trait, baseline locomotor activity is likely to be influenced by multiple genes with a strong interactive component. Activity levels of 254 male mice from 24 lines of the BXD recombinant inbred panel were measured in a home-cage task and in silico interval mapping was carried out using WebQTL (www.genenetwork.org). A chromosome 11 QTL was detected at 70cM which includes the Aanat gene. The AA-NAT enzyme is involved in melatonin synthesis which functions as a hormonal message for the circadian rhythm in mammals. C57BL/6J mice are known to be melatonin deficient due to a G?A point mutation in Aanat. However little was known about the genotype of this allele in most laboratory inbred mouse strains, although it is known that DBA/2J mice have low but detectable melatonin levels. Therefore, this point mutation in Aanat was genotyped across 48 inbred strains and a GG genotype was identified in DBA/2J, supporting the hypothesis that the segregation between C57BL/6J and DBA/2J strains at this allele could contribute to the baseline activity QTL detected at Chr11. Association between this mutation in Aanat and baseline activity was further explored using an outbred heterogeneous stock (HS) mice.
Social, Genetic and Developmental Psychiatry Centre, Institute of Psychiatry, King's College London, UK.

POSTER SESSION I Abstracts

Generalisation of conditioned fear in mice deficient for the neural cell adhesion molecule NCAM.
A Albrecht, J Bergado, H-C Pape, O Stork
In classical fear-conditioning animals learn quickly to associate a previously neutral sensory stimulus (CS+) like a tone with an aversive stimulus (US) such as foot shock. Although this paradigm is commonly used for studying processes of emotional memories, little is known about mechanisms underlying generalisation. Generalisation, describing a transfer of the conditioned response to neutral test stimuli (CS-) or to the training environment (context), occurs after intensive training with highly stressful US (overtraining). Using an auditory fear-conditioning paradigm we began to investigate molecular aspects of such generalisation in mice deficient for the neural cell adhesion molecule (NCAM-/- mice). NCAM is a glycoprotein of the immunoglobulin superfamily which plays important roles in both, CNS development and synaptic plasticity. NCAM-/- mice display deficits in emotional behaviour (increased aggression and anxiety) and memory formation, but pre-exposure to context and CS- could overcome previously observed deficits in cued and contextual fear conditioning. In our study we found a comparable increase of defensive behavior towards the CS- (intramodal generalisation) in overtrained NCAM-/- and NCAM +/+ animals. However, while overtrained NCAM+/+ mice also increased their defensive response (risk assessment and freezing) to the conditioning context, their NCAM-/- littermates showed no such generalisation. In according with findings from previous studies indicating a stress-regulated NCAM-expression, our data suggest the critical involvement of NCAM-mediated cellular processes in stress-dependent modulation of emotional memory, in particular, its generalisation to contextual background information. This deficit was correlated with changes of synchronised network activities in the amygdalo-hippocampal pathway of fear conditioned NCAM-/- mice.

Institut für Physiologie, Medizinische Fakultät, Otto-von-Guericke Universität Magdeburg, Leipziger Str. 44, D-39120 Magdeburg, Germany. Supported by Deutsche Forschungsgemeinschaft (SFB 426 TP B7; Leibniz-Programm).

Characterizing the phenotype of a transgenic mouse model overexpressing β4, α3 and α5 nicotinic achetylcholine receptor subunits.

X Gallego, M Dierssen

Nicotinic acetylcholine receptors (nAChRs), are members of a superfamily of ligand-gated ion channels that mediate fast signal transmission at synapses. The nAChRs are thought to be (hetero) pentamers composed of homologous subunits. Nictonic receptors have been related to anxiety responses, mainly those containing beta-4 subunits. The beta-4 subunit is included in a gene cluster, next to the alpha-3 and alpha-5 subunits located on human chromosome 15 in the region 15q24. Previous studies in humans suggested that segmental duplications of this chromosomal region could be involved in panic disorder. To explore the effect of the overexpression of these subunits in anxiety we generated a transgenic mouse model. Two founder mice overexpressing CHRNB4, CHRNA3 and CHRNA5 were produced after microinjection into fertilized B6SJL mouse oocytes, using a construct obtained by digestion of the BAC RP11-335K5 (AC067863). Expression of the transgene was determined by RT-PCR analysis and the distribution pattern of the Chrnb4, Chrna3 and Chrna5 subunits was analyzed by immunohistochemistry. Transgenic mice were viable and showed no alterations in fertility, perinatal mortality or somatometric parameters. The phenotypic characterization using a battery of tests including sensorimotor and neurologic tests showed no differences in sensory or neurological parameters. Activity and anxiety-related behavior tests, such as the Open Field, Plus-Maze and Fear Conditioning are in progress.

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Understanding the pathogenetic mechanisms of Panic Disorder: consequences of NTRK3 overexpression on fear circuit.

I Sahun, M Dierssen

It has been suggested that neurotrophic factors may participate in the pathophysiology of anxiety disorders. The neurotrophin type 3-receptor (NTRK3) is expressed in the locus coeruleus (LC) and its ligand, NT-3, has a role as a survival factor for noradrenergic neurons. The main symptom of persons with anxiety/panic disorders is their inability to correctly identify the fear-related information that may depend on altered responsiveness of fear circuits and inappropriate hyper-responsiveness of the NA system is observed in panic disorder (PD) patients. In a first series of experiments we performed a detailed neuromorphometrical analysis of the amygdala and hippocampus by means of stereological techniques. We showed a tendency to an increased cellular density in all hippocampal subregions in TgNTRK3 and a significant increa! se in the density and number of cells of the basolateral amygdala, along with a tendency to increase its volume. To analyze the possible functional consequences of these structural modifications, we used neurobehavioral paradigms like the Fear Conditioning (FC) and Morris Water Maze (MWM). We showed an increased sensitivity to context and non-context FC in TgNTRK3 24 hours after training and 1 week after the first test. Nevertheless, in the hippocampal-dependent MWM task, no differences were shown in escape latencies across acquisition sessions between genotypes. We propose that the hypersensitive fear system in PD depends on hypertrophy of key regions of the fear circuit, as the basolateral amygdala that significantly disrupts the formation of emotional memories. This increase in emotional memory-efficacy may depend on an increased release of norepinephrine from LC evoked by the foot-shock during the training phase of FC. The results suggest the involvement of NTRK3 in anxiety leading to a neurotrophic hypothesis for the pathogenesis of PD.

Neurobehavioural Analysis of Genetically Modified Mice. Genes and Disease Program, Genomic Regulation Center (CRG), Barcelona, Spain. This work was supported by SAF2001-1231 and DURSI (Generalitat de Catalunya).

Somatostatin is involved in contextual fear memory formation.
C Stoppel1, O Stork1, H-C Pape1,2
Somatostatin (SOM) has been shown to be involved in aversive learning and other memory tasks. However, the role of SOM in fear conditioning has not been elucidated so far. In this study we investigated the effects of genetically and pharmacological induced SOM deficiency onto cued and contextual fear conditioning in mice. After conditioning to an auditory cue (CS+) SOM null mutants (SOM-/- mice) were scored for freezing onto background-context, CS+ and an unconditioned tone (CS-). SOM-/- mice displayed selectively decreased freezing to background-context, while no differences in response to CS+ and CS- could be observed. To further investigate the involvement of SOM in fear memory formation, foreground contextual fear conditioning was performed. As in cued fear conditioning freezing to the context was reduced in SOM-/- mice. To test whether the observations in SOM-/- mice are due to acute involvement of SOM in fear memory formation rather than to developmental deficits, pharmacological depletion of SOM was performed by intraperitoneal infusion of cysteamine (50 or 150 mg/kg). On one hand, application 4h pre-training led to specifically decreased freezing to background-context, thus phenocopying the null mutant phenotype. This effect was not observed upon injection to SOM-/- mice. On the other hand, cysteamine infusion 10min post-training led to a generalised fear-response to context, CS+ and CS- in C57Bl6 mice and SOM-/- mice, too. Therefore this finding is supposed to be rather unspecific and not dependent upon somatostatinergic mechanisms. Together, our results indicate that SOM plays a critical role in memorising contextual information interconnected with a threatening experience.
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Role of Dyrk1A in neuromotor development of CNS: implications in Downs Syndrome.
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Motor deficits neuromotor development delay are among the most frequent impairments in Downs syndrome (DS), but their neuropathological and molecular bases remain elusive. Here we investigate the motor profile of transgenic mice overexpressing Dyrk1A, TgDyrk1a, a candidate gene hypothesized to cause some of the neurological defects associated with DS. DYRK1A is expressed in the cerebellum and functionally related structures, most brainstem motor nuclei and spinal cord, supporting a role for Dyrk1A in the neural motor pathway involved in neuromotor development during the postnatal period and in controlling motor function in the adult. In previous studies we demonstrated a persistence of immature locomotor patterns, delayed walking activity and retarded general psychomotor development. Here we extended our previous neurodevelopmental screening in TgDyrk1A by using different neurological and behavioral tests. Our results support the notion of a specific motor alteration that impedes the correct performance of coordination/postural adjustment tests. Anxiety-like behavior is detected thus confirming results in adult mice. We also studied the involvement of Dyrk1A in the development of motor cholinergic nuclei in the spinal cord (specially in the ventral horn). Transitory Dyrk1A immunostaining was observed in neurons of reticular formation at PD7 whereas at later stages this pattern was shifted to cholinergic (ChAT positive) neurons in the facial nucleus. The characterization of this Dyrk1A positive population in the reticular formation demonstrated that GABAergic neurons were mainly coexpressing Dyrk1A. At P14 and in the adult Dyrk1A was observed in cholinegic neurons of the facial nucleus. In the ventral horn of the spinal cord expression of Dyrk1A in motoneurons started earlier ion development (PD10) and persisted through adulthood. We propose, that Dyrk1A may play a role in the development of the motor system with functional consequences arising from its overexpression.
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Protective effect of green tea on brain alterations in murine models overexpressing dyrk1a.

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A transgenic mouse containing a 500kb human YAC clone (152F7) (Smith et al.) was constructed to model the overexpression of genes from the Down syndrome chromosomal region-1 on HSA21. This model presents two interesting phenotypic modifications: a learning and motor impairment and a volumic increase of some parts of the brain with a more pronounced effect on the thalamic-hypothalamic region as evidenced through MRI experiments. The transgenomic fragment contains five genes among which is dyrk1a, a serine threonine kinase, ortholog of drosophila minibrain. QPCR experiments have shown that the transgene is present in one copy and induces a 1.5 increase of the expression level of dyrk1a in the cerebrum. Crossing of tg152F7 with dyrk1a (+/-) heterozygote produces four genotypes [wt, tg, (+/-), tg (+/-)], analysis of which shows that the brain phenotypes are strongly correlated to dyrk1a gene copy number and to dyrk1a expression level. As a consequence any drug acting upon dyrk1a level or dyrk1a activity should also act upon the phenotype. This kinase has been shown to be strongly inhibited in vitro by epigallocatechin gallate (EGCG) a major component of green tea (Bain et al). We have studied the effect of a green tea diet on the brain volumic increase of YAC transgenic animals as compared to wild type: green tea administered orally during gestation and postnatally can reverse brain phenotypic changes induced by dyrk1a overexpression.

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The cerebral cortex in Down syndrome murine models.

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Down syndrome results in neuropathological alteration of cerebral cortex anatomy. However, the pathogenetic background of cortical irregularities is presently not known. The dual-specificity tyrosine-regulated kinase DYRK1A gene maps to the chromosomal segment HSA21q22.2 in the Down syndrome critical region (DSCR), and is believed to be involved in some neurological deficits of DS. It has been demonstrated previously that Dyrk1A has dosage-dependent effects on cognition and behavior. In the present work we analysed the microstructure of cortical circuitry in Dyrk1A murine models and control littermates by intracellular injection of Lucifer Yellow in neurons in fixed cortical tissue. We found that pyramidal cells in Dyrk1A mice were considerably smaller, less branched and less spinous than those sampled from control littermates. These results suggest that Dyrk1A is involved in the determination of the size and complexity of pyramidal cells, and thus, in their capability to integrate information.

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Dopaminergic system is altered in a murine model with haploinsufficiency of Dyrk1A.

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Dyrk1A has been considered a good candidate gene for the Down syndrome (DS) phenotypic abnormalities due to its localization in the DS critical region of human chromosome 21 and its involvement in central nervous system development. To study the in vivo function of Dyrk1A, we have characterized mice with loss of function of Dyrk1A. Since the null mutants died during midgestation, we used heterozygous mice (Dyrk1A+/-). Dyrk1A+/- mice show a hypodopaminergic phenotype characterized by hypoactivity in the wheel running test and an aggravation of motor alterations at advanced ages. Dyrk1A +/- mice are less sensitive to the cataleptic effect of dopaminergic D1 and D2 antagonists in the bar test. Moreover, the amphetamine-induced release of dopamine in the striatum was lower in Dyrk1a+/- than in control mice, as assessed by microdialysis in freely moving mice. This result was confirmed by in vivo PET studies in which the incorporation of 18-FDG was lower in Dyrk1A+/- mice after amphetamine injection. In the substantia nigra, stereological methods revealed an increased dopamine neuronal density but a reduced volume and total cell number in Dyrk1A+/-, compared with control mice. Our data support the hypothesis that Dyrk1A may participate in the DS motor phenotype through an alteration of the dopaminergic system.

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Mouse models of Down syndrome trisomic for all human chromosome 21 syntenic regions.

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Down syndrome consists of a constellation of developmental abnormalities that occur in 1 of 800-1000 live births. The underlying cause of this disorder is trisomy 21. Modeling Down syndrome in mice is feasible because of the syntenic conservation between human chromosome 21 and 3 genomic segments on mouse chromosomes 10, 16, and 17. The Ts65Dn strain is currently the best mouse model, and it is trisomic for about 14.2-Mb of the orthologous region on mouse chromosome 16. Functional annotation has indicated that the genes encoding components of several cellular pathways are located in different orthologous mouse chromosomal regions. Therefore, the ideal model should be trisomic for all three orthologous mouse genomic regions. To establish such a model, we have engineered a 2.5-Mb duplication in the orthologous region on mouse chromosome 10, (Ts10)1, by using Cre/loxP-mediated chromosome engineering. The endpoints of the duplication are located in positions that are immediately adjacent to the orthologous region. Ts(10)1 mice were viable and fertile. In order to assess the phenotypic impact of Ts(10)1, this strain is being crossed to Ts65Dn. Furthermore, 22.9-Mb and 1.2-Mb duplications in the orthologous regions of mouse chromosome 16 and 17, respectively, are being developed in our laboratory.

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Resistance to ß-amyloid aggregation and Aß-induced cell death in mice lacking GD3 synthase.

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Studies in vesicle preparations have shown that gangliosides are necessary for ß-amyloid (Aß) binding and aggregation. In addition, the ganglioside GD3, which is increased in Alzheimer's disease, has been shown to be necessary for Aß-induced apoptosis. Knocking out the GD3 synthase (GD3S) gene effectively eliminates half of the major brain gangliosides, including GD3. Knockout mice lacking GD3S are viable, grossly normal, and have a normal lifespan. The present studies investigated the relationship between membrane gangliosides, Aß aggregation, and cell death, using mice null for GD3S. Primary neuronal and astrocyte cultures were established from 1-day-old mouse pups lacking GD3S, or wild-type controls. Aß binding was visualized using fluorescent-labeled antibodies to Aß after application of exogenous Aß 1-40. DNA damage was assessed using triphosphate-biotin nick end labeling (TUNEL) after 24-hr. incubation with Aß 1-42. GD3S-/- mice and wild-type littermate controls were assessed on a battery of memory tests at young and old ages. In astrocytes from GD3S knockout mice, there was a gene-dose-dependent reduction in exogenous Aß binding, with a greater than 50% reduction in homozygous knockout mice. Neurons from GD3S null mice were completely resistant to apoptosis after exposure to 10 µM Aß 1-42, a concentration that induced widespread cell death in wild-type neurons. Behavioral studies showed that GD3S-/- mice were normal on tests of locomotor activity, balance and coordination, anxiety, and memory. These results suggest that strategic reduction of gangliosides may be a novel therapeutic strategy to combat both the formation of amyloid plaques and the widespread cell death that afflicts Alzheimer's patients.

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PCP4 (PEP-19) : modelization of overexpression.

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PCP4 belongs to a family of proteins involved in calcium transduction signals. It binds calmodulin as neuromodulin and neurogranin via an IQ motif, but it is not regulated by PKC. PCP4 has been shown to modulate calmodulin targets such as CaMKII (Johanson 2000) and nNOS, which are involved in the transduction of apoptotic signal and in neuronal plasticity. More recently, Putkey (2003) showed that PCP4 highly accelerates both the association and dissociation of calcium from calmodulin. The gene is localised on human chromosome 21 and on mouse chromosome 16. Its expression was first described post-natally and in adult in many brain regions with a neuronal pattern. Our work showed that *pcp4* is expressed very early in development in ectoderm and neuroectoderm comprising neural crest derived cells. Using immunohistochemistry with a specific antibody, we compared the mRNA and protein patterns, which, in the brain, show to be identical even for the relative levels. Its pre- and post-synaptic localisation in neurons was confirmed inside the hippocampus. To evaluate the direct consequences of its overexpression, which has been confirmed in partial ts16 models of human trisomy 21, we have constructed an animal model with a human transgene containing all its own regulatory sequences. The phenotypes of this model will be presented.

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DSCR1(Adapt78) and calcipressin 1 inhibit calcineurin and induce Gsk-3: possible role in brain functions.

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DSCR1(Adapt78) is a multiple stresses inducible gene. The DSCR1(Adapt78) protein, called calcipressin 1, binds to calcineurin, a serine/threonine phosphatase (PP2B), and inhibits it's activity 3-6. Here we demonstrate that calcipressin 1 also induces GSK-3, a kinase that can antogonize the action of calcineurin. Both calcineurin and GSK-3 have been demonstrated as a critical regulators of synaptic plasticity and process of learning and memory. They are key regulators of tau phosphorylation, and of NFATs (nuclear factors of activated T cells). Hyperphosphorylation of the tau protein can lead to neurodegeneration, such as that observed in Alzheimer disease. Inhibition of NFATs can lead to heart defects, skeletal muscle abnormalities and immune system deficiencies. Interestingly, all of these pathologies are observed in Down syndrome. Since we also find that DSCR1(Adapt78) is overexpessed in tissues affected by Alzheimer disease and Down syndrome, our data provide a possible mechanistic link between DSCR1(Adapt78) overexpression and the pathologies observed in these .

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Spatial and non-spatial Barnes-maze learning in APP+PSEN1 double-transgenic and wild-type mice.

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The Barnes maze is used to assess spatial learning in rodents and, because it does not involve swimming, is considered to be less stressful than the Morris water maze. The Barnes maze and water maze share two standard conditions; a spatially fixed escape position without proximate cues, and a spatially variable escape position associated with a proximate cue. The standard procedure involves training animals to locate the hidden, spatially fixed target before testing on the non-spatial cued-target condition. The cued target condition is used to evaluate non-spatial learning. In the current study we assessed the relative importance of spatial (extra-maze) and non-spatial (intra-maze) cues, as well as more general rule- and context-learning in the Barnes maze. In Experiment 1, we used a double transgenic mouse model of Alzheimer's disease (APP/PSEN1) to evaluate learning on the Barnes maze. Mutant and wild-type B6C3F1/J mice were trained for 5 consecutive days on both cued and non-cued conditions, in counterbalanced order. Transgenic mice showed a learning deficit only in the hidden target treatment, and only when that treatment was preceded by the cued version of the task. In Experiment 2, we trained wild-type mice on one of 4 experimental conditions for 5 consecutive days. In a hidden target constant position (HTCP) condition, the escape box location was always in the same place relative to distal room cues. In a cued target variable position (CTVP) condition an unambiguous cue

marked the position of the escape hole. A cued target constant position (CTCP) condition and a hidden target variable position (HTVP) condition, in which the escape hole position was randomly varied between trials, were also included. Our results indicate that mice are able to locate the escape box and solve the Barnes maze regardless of the extra-maze (distal) and intra-maze (proximate) cues available to them, and that they use both spatial and non-spatial strategies with near equal proficiency and frequency by the end of training. We conclude that, although the Barnes maze clearly requires spatial skills, it can be solved efficiently using non-spatial strategies that depend more on context and rule learning than on learned spatial relationships.
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Genetic dissection of autism-related social behavior in the mouse.
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Autism is a uniquely human disorder comprised of three major behavioral components: social deficits, repetitive/ritualistic behavior and communication abnormalities. There is a wide range of severity observed in autistic patients as well as the milder, non-clinical manifestation of the Broader Autism Phenotype (BAP) in family members. Just as humans can span the range of social interaction phenotypes, from autistic to BAP to reserved to outgoing, various inbred lines of mice exhibit a range of different behaviors and social preferences. We are correlating gene expression profiling with social behaviors of ten inbred strains in order to begin to dissect the complex genetics of autism. Social behaviors are being modeled by determining the preference of mice to spend time around and interact with novel mice. We observe that sociability varies from strain to strain, indicating a strong genetic component. In order to profile the expression of genes associated with autism-like behaviors, microarray analyses are being conducted on seven brain regions implicated in autism or social behavior. Data on strain-dependent gene expression will be presented. In this way, we hope to begin to dissect the complex genetics of social behaviors and autism.
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The study of prevalent beta-thalassemia gene mutation in thalassemic patients of Khouzestan province, Southwest Iran.
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Beta-thalassemia is one of the most common inherited single gene disorders in Iran. More than two million carriers of beta-thalassemia are living in Iran. Since the Iranian population is a mixture of different ethnic groups, it was necessary to determine the frequency and distribution of mutation in the different parts of the country. For this purpose, we studied 200 affected patients who were residents of Khouzestan province, Iran. The technique we applied in this research was Reverse Dot Blot. We detected 78/4% beta-thalassemia mutations in the studied chromosomes. Codon 36/37 mutation was the predominant mutation found in our study (14/7%). IVS-I-110(14/2%), IVS-II-1(6/9%), Codon8(6/5%) and Codon5 (5/2%) were the other most common mutations respectively. In our study 21/6% of mutations were undiagnosed with reverse dot blot technique.

Sleep behavior in mice modeling Angelman syndrome.
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Background: Angelman syndrome (AS) is characterized by neurodevelopmental impairment, electroencephalographic abnormalities and sleep disturbances, associated with functional deficit of UBE3A gene. Different mechanisms of UBE3A inactivation correlate with clinical phenotypes of varying severity. Methods: Ube3a-maternal deficient mice [Ube3a (m-/p+)] were generated in a

C57Bl/6J background. This study compares changes occurring in cortical EEG and sleep-wake state architecture in adult Ube3a (m-/p+) mice compared with age-matched Ube3a (m+/p+) mice, under baseline conditions or after a 4h sleep deprivation (SD). Results: Ube3a m-/p+ mice exhibited: decreased slow wave sleep (SWS) amount at the expense of the waking state (W) at the dark/light transitions; increased SWS and W episode numbers; and drastic rapid eye movement sleep (REMS) deterioration over 24 h [amount: - 44 %; episode duration: - 46 %; episode number: -40%; theta peak frequency (TPF) acceleration: 7.6 Hz vs 7.0 Hz in control mice]. Characteristic discharges during W and SWS are also observed. Following recovery subsequent to SD, Ube3a m-/p+ mice exhibited no rebound in slow wave activity (SWA) (+89% in control mice) but a slight (20%) rebound in REMS amount. In spontaneous wakefulness, hippocampal and cortical c-fos and zif-268 expressions were up regulated. Conclusions: This first study opens a new path toward further insight into molecular mechanisms implicated in the communication of memory traces in relation with the sleep-related disturbances observed in AS subjects.
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Impaired behavioral control and altered processing of spatial information in mice deficient for the X-chromosomal mental retardation gene Arhgef6.
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The ARHGEF6 gene (also known as alphaPIX or Cool-2), encodes a protein with homology to guanine nucleotide exchange factors (GEF) for Rho GTPases. Mutations in this gene have been recently identified in human patients with X-linked mental retardation (Kutsche et al. Nat Genet 26:247-250, 2000). We report the cognitive and behavioral characterization of male mice carrying a targeted deletion of Arhgef6. These mice are healthy and have normal appearance. They were evaluated in a water-maze place navigation task, in a spatial working memory procedure on the 8-arm radial maze, as well as in a set of tests assessing locomotor activity, anxiety, exploratory behavior and their reaction to novel stimuli. In the water maze, they produced navigation errors and showed spatial perseverance when required to adapt to a changed goal position. Their reaction to a novel stimulus within in a familiar environment was clearly disinhibited. Spatial reference and working memory as such, as well as day to day habituation to a novel arena were intact. We found no indication of altered basal activity, anxiety, or abnormal adaptation to stressful situations. Moreover, detailed analysis of locomotion and swim patterns did not reveal neurological deficits. In summary, Arhgef6 null mice displayed specific behavioral alterations suggesting deficient behavioral control and altered processing of spatial information. The behavioral profile of Arhgef6 null mice is reminiscent of phenotypic changes observed in other mouse models, with impaired function of the hippocampus and connected cortical regions.
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Suppression of two major Fragile X Syndrome mouse model phenotypes by the mGluR5 antagonist MPEP.
QJ Yan, M Rammal, M Tranfaglia, RP Bauchwitz
Fragile X Syndrome is the most common form of inherited mental retardation worldwide. A Fragile X mouse model, fmr1tm1Cgr, with a disruption in the X-linked Fmr1 gene, has three substantial deficits observed in several strains 1) sensitivity to audiogenic seizures (AGS), 2) tendency to spend significantly more time in the center of an open field, and 3) enlarged testes. Alterations in metabotropic glutamate receptor group I signaling were previously identified in the fmr1tm1Cgr mouse. In this study, we examined the effect of MPEP, an antagonist of the group I metabotropic glutamate receptor mGluR5, on audiogenic seizures and open field activity of fmr1tm1Cgr mice. Genetic analysis revealed synergistic reactions between fmr1tm1Cgr and inbred AGS alleles. In addition, AGS sensitivity due to the fmr1tm1Cgr allele was restricted during development. Examination of phenotypes combining mGluR5 inhibition and Fmr1 mutation indicated that absence of FMRP may affect mGluR5 signaling through indirect as well as direct pathways. All

strains of fmr1tm1Cgr mice tested, (FVB/NJ, C57BL/6J, and an F1 hybrid of the two), had a more excitable AGS pathway than wild type, and consequently required more MPEP to achieve seizure suppression. In open field tests, MPEP reduced fmr1tm1Cgr center field behavior to one indistinguishable from wild type. Therefore, modulation of mGluR5 signaling may allow amelioration of symptoms of Fragile X Syndrome.
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Effects of choline supplementation on the behavioral phenotype of MeCP2 mutant mice.
R Yang, N Nag, J Berger-Sweeney
Mutations in the X-linked gene encoding MeCP2 are responsible for approximately 80% of all cases of Rett Syndrome (RTT): a neurodevelopmental disorder affecting nearly 1 in 10,000-22,000 females and often lethal for males, if not severely debilitating. Morphological analyses of the brains of RTT patients show abnormal development of the cholinergic basal forebrain, decreased cholinergic markers such as Choline acetyltransferase (ChAT), and atrophy in both the cerebrum and cerebellum. In a number of animal models of various neurological diseases, choline supplementation during prenatal and postnatal periods of development has been shown to facilitate an improvement in memory. This is thought to be accomplished through a number of mechanisms including increasing and enhancing acetylcholine transmission, altering cell size and distribution, and potentially affecting synaptogenesis. We hypothesize that choline supplementation of MeCP2 null mice, an animal model of RTT, will rescue the cognitive deficits seen in these mutants. To examine this hypothesis, MeCP2 mutants, and their wild type litter mates, were supplemented with either 25mM choline / 50mM saccharin (test) or 50mM saccharin alone (control) from embryonic day (ED) 10 to weaning (3 weeks). At approximately 4 weeks, mice were subjected to the following behavioral tasks 1) neurological battery to examine basic reflexes, 2) locomotor activity and 3) rotor rod for to assess motor coordination; and 4) cued and contextual fear conditioning, to assess associative learning. Our results may provide insight into a potential pharmaceutical intervention to reverse the impairments observed in RTT.
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Regulation of hippocampal long-term potentiation and hippocampus-dependent spatial learning by D-serine and glycine.
V Labrie, S Duffy, J Roder
The NR1 subunit of the NMDA receptor contains a high affinity glycine-binding site at which either glycine or D-serine can act as an obligatory co-agonist. We examined the unique roles of D-serine and glycine in modulating NMDA-dependent hippocampal long-term potentiation (LTP) and hippocampus-dependent spatial learning. In hippocampal slices from adult C57BL/6J mice, the D-serine catabolic enzyme D-amino acid oxidase reduced LTP at Schaffer collateral-CA1 synapses. Similarly, L-serine O-sulfate, an inhibitor of the D-serine synthetase serine racemase, inhibited LTP, indicating that D-serine is the dominant physiological co-agonist in CA1. A high concentration of exogenous D-serine (100ìM) did not alter LTP, indicating that synaptic D-serine was saturating during LTP induction. The Grin1D481N mutation confers a 4-5 fold lower glycine affinity, and Grin1D481N/ D481N mice demonstrated a deficiency in LTP that was rescued by exogenous D-serine, but not by t! he GLYT-1 blocker ALX-5407. Pre-injection of D-serine did not alter spatial learning in wild-type mice (Grin1+/+) as measured by the reference memory version of the Morris water maze task. D-serine did rescue spatial learning in Grin1D481N/ D481N mice, while ALX-5407 was without effect. Spatial learning was correlated with hippocampal LTP under all conditions. We propose that exogenous D-serine did not alter hippocampus-dependent learning in Grin+/+ mice because the NR1 glycine site was saturated by D-serine and so NMDA-dependent plasticity was unaffected. Alternatively, exogenous D-serine rescued poor spatial learning in Grin1D481N/ D481N mice by restoring NMDA-dependent plasticity in the hippocampus. D-serine is an effective cognitive enhancer when D-serine is deficient, as may occur in schizophrenia.
Society Samuel Lunenfeld Research Institute, Mount Sinai Hospital.

Differences in hippocampal gene expression in HS mice selected for Low, Middle, and High performance on cognitive tasks as measured via GeneChip array and real-time PCR.

MJ Parsons, JL Paya-Cano, C Fernandes, L Liu, LC Schalkwyk

The genetic components influencing individual differences in cognitive ability is likely to be complex. Heterogeneous Stock (HS) mice display marked individual differences in behaviour as well as providing greater genetic variability for behaviour genetics studies, therefore it is possible to select individuals for quantitative variations in the trait of interest Based on composite scores derived from a battery of cognitive tasks, we selected three groups of mice low (n=27), mid (n=27) and high (n=27) cognitive ability that represented the lower, middle and upper ten percentile from a population of 270 HS mice. We examined hippocampal gene expression profiles using the Affymetrix MOE430A GeneChip array across these three groups. Twenty-seven genes showed significant differences in expression between the three groups. In order to verify these findings within the individual samples the relative gene expression for three of the strongest candidate genes (stk25, ttr, vamp3) was assayed using a Taqman based expression assay. Preliminary analysis of the Taqman data suggests that there is a statistical trend towards differential expression in at least one of these three genes ($p<0.06$), though further analyses is necessary. From the results presented here, we conclude that at least some of the differential expression from the GeneChip profiling represents real differences in gene expression caused by differences in cognitive performance.

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Environmental enrichment behavioural effects in mice, selected for large and small brain weight.

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Two experiments were performed with female mice from lines selected for large and small brain weight (LB, SB) (generations F10 and F11). In both experiments mice lived for 3 months either in "enriched" or in standard laboratory environment. After that they were tested in open field, in closed plus-maze and slip funnel. Their learning and problem solving capacities were tested too. Interstrain brain weight differences were significant (ANOVA), while no differences in this index were found between "standard cage" and "enriched" groups. Environmental enrichment resulted in the increase of fear reactions (freezing) in mice of both strains (open-field data), while exploration strategy in closed plus-maze did nor differ. The scores of correct task solution ratios in the extrapolation task (which evaluates the ability to extrapolate the direction of food stimulus movement) were significantly higher in LB "enriched" mice in comparison to the LB standard environment animals and to both groups of SB line. The simplified water maze test (small pool - 60 x 60 cm, with one day 6 trials session with permanent platform location) revealed the superiority of mice of "enriched" groups - both LB and SB - they performed more straight swim trajectories towards platform. T-maze learning (food reinforcement) were performed with shorter latencies by "enriched" LBs. Thus 3 months experiences in large cages which provides physical exercises and perception enrichment resulted in the increase of cognitive abilities which was differentially expressed in strains differing by brain weight. As environmental enrichment is reported to intensify the adult brain neurogenesis these data could indicate that genetic brain weight differences could influence the intensity of such effect.

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Novel candidate genes for cognitive performance using outbred heterogeneous stock (HS) mice and BXD Recombinant Inbred (RI) strains.

JL Paya-Cano, C Fernandes, L Liu, MJ Parsons, R Plomin, LC Schalkwyk

Genome-wide studies of gene expression as assessed by DNA microarray technology are beginning to be applied to the study of complex traits in mice, including cognitive processes such as learning and memory. We have tested a large sample of outbred heterogeneous stock mice (HS), a set of BXD recombinant inbred strains (BXD RI), and two inbred strains (C57BL/6J and DBA2/J) in a battery of diverse behavioural tests assessing activity, exploration, anxiety, learning,

memory, and problem solving. Our research has focused in three main areas: 1) We have used HS mice to suggest novel candidate genes for cognition by examining hippocampal gene expression profiles associated with low, mid and high levels of performance in a set of cognitive tasks; 2) Making use of WebQTL, we tentatively suggest quantitative trait loci (QTL) for a subset of behavioural measures and other morphological traits such as weights of brain, hippocampus and cerebellum in the BXD RI mice; and finally 3) we have performed genetic correlation analyses between these measures with other phenotypic traits including total brain gene expression from WebQTL. Overall, the results presented here may provide additional insight into the genetic mechanisms of cognition in mouse, including mechanisms involving novel candidate genes.
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Prepulse inhibition of the acoustic startle response increases during repetitive testing.
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The acoustic startle response (ASR) is a general coordinated muscle contraction elicited by loud acoustic stimuli. A non-startling acoustic prepulse administered before the startle-eliciting stimulus elicits two opposite and independent processes: either an ASR decrease (prepulse inhibition, PPI) or an ASR increase (prepulse facilitation, PPF). The respective strengths of PPI and PPF mainly depend on the interpulse interval (IPI) between prepulse and startle-eliciting stimulus. In mice, PPI overbalances PPF at an IPI of 50 ms leading to an overall ASR inhibition. This inhibition increases when it is repetitively elicited over several days. The reason for this may be either an increase in PPI or a decrease in PPF. Here we show that the PPI increase over days is not caused by a PPF decrease. In C3H mice, PPI increased over days while PPF remained constant. In C57 mice almost no PPF occurred (which we believe to be typical for this strain), while PPI increased. We further show that the full amount of the PPI increase over days is only produced if the prepulse and the startle-eliciting stimulus are presented in a contingent manner. We trained 5 different groups of C57 mice on 4 days: In 1 group the two stimuli were presented contingently, while in 4 control groups only (i) the startle-eliciting stimulus, (ii) the prepulse, (iii) the experimental context were presented or (iv) startle stimulus and prepulse were given in a non-contingent manner. The amount of PPI was then compared between the groups on days 5-8, on which all groups got contingent pairs of prepulses and startle stimuli. In the 4 control groups PPI increased, but this increase was in all groups only about half of the amount of the PPI-increase of the contingent group. From this we conclude that the PPI increase during repetitive testing is due to a learning process of the association between prepulse and startle stimulus, and that this process is facilitated by previous presentation of context or stimuli.
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POSTER SESSION II Abstracts

Automated behavioral phenotyping of mutant mice: how to handle large data sets.
BM Spruijt, L de Visser, P Kuurman
The increasing numbers of genetically modified animals to be characterized prompted us to develop a tool for observing and analyzing challenge-induced and baseline behavior. So far test batteries have two main disadvantages: (1) behavior is induced in a novel environment and the limited observation time excludes habituation, baseline levels, circadian rhythmicity, etc. and (2) most tests have a specific focus on one motivational system, e.g. exploration, anxiety, etc., and, subsequently, measure a limited number of parameters. A functional interpretation of gene effects on behavior requires an ethological approach allowing an ongoing interaction of various motivational systems. We present a newly developed method for behavioral phenotyping, which consists of an enriched home cage (PhenoTyper, Noldus Information Technology, Wageningen, The Netherlands) equipped with hardware and software allowing automated observation over

days. The interaction of multiple motivational systems is due to the presence of a shelter, food, water and the programmable illumination of each zone. Limitations of this approach resides not in the measurement of behavior, but rather to what extent large sets of data can be reduced to a functional description permitting the detection of changes in complex behaviors of vast numbers of animals. Starting off with defining a comprehensive ethogram, mathematical modeling will provide the read-out parameters of complex behavioral categories, e.g. activity. These read-out parameters, representing their distribution in both time and space are appropriate for being subjected to statistical procedures such as principal component analysis. This will result in the reduction of data into biological relevant categories, which will in turn form the key elements of a behavioural profile. In short, a complex test requires complex statistics to have the full benefit of this approach. The fact that we collected data over days contributes significantly to the power of such factor and subsequent pattern analysis.
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Approaching standardization of behavioral testing in mice: inter-laboratory strain comparisons of manually conducted tests versus automated procedures using IntelliCage.
A Vyssotski, D Vyssotski, O Litvin, AE Rau, S Morf, H Würbel, RM Nitsch, DP Wolfer, H-P Lipp
Even by using stringent standardization criteria, behavioral tests of mice often reveal discrepancies between laboratories, especially in tests depending on spontaneous activity or involving fear and exploration. In a recent, carefully standardized multi-lab study, we noted that enriched environment did not alter the strain rank order in the open-field, 0-maze, water maze and object exploration. On the other hand, the comparison of absolute values revealed several significant differences between laboratories, or strain-by-laboratory interactions. A subset of 47 female mice was also tested in fully automated cages (INTELLICAGES) located in different animal facilities of the university of Zurich, each cage housing 12 transponder-tagged mice from different strains. The system measured continually visits of drinking sides (4 per cage), development of place preference and reversal, and other parameters, usually over periods between 24-48h. Thus far, INTELLICAGE always showed statistically indistinguishable behavioral scores from the two locations, but also revealed significant strain differences in several yet not all measures. This implies (i) that much of the previously observed differences between laboratories must be attributed to human handling and uncontrollable environmental variations in test set-ups, and (ii) that optimal standardization and comparability is best achieved by the use of automated procedures.
Division of Neuroanatomy and Behavior, Institute of Anatomy and Psychiatry Research Department, University Hospital Zürich, University of Zürich, Switzerland, and Institute of Veterinary Physiology, University Giessen, Germany. This work was supported by the Swiss National Foundation and the NCCR Neural Plasticity and Repair.

Effects of various factors on the results of a comprehensive behavioral test battery for genetically engineered mice: a factor analytic study.
N Yamasaki1,2, K Tanda1,3, A Yamada1, K Toyama1, T Miyakawa1
We have been using a behavioral test battery to reveal unknown phenotypes of genetically engineered mice. Our behavioral test battery includes general health and neurological screen, light/dark transition, open field test, elevated plus maze, social interaction test, rotarod, prepulse inhibition, Porsolt forced swim test, tail suspension test, 8-arm radial maze, cued and contextual fear conditioning, and 24-hour social interaction test in home cage. For the adequate experimental design and interpretation of data, it is essential to know experimental variables which may potentially influence results, and various kinds of factors which underlie many indices measured in the tests. In this study, we investigated the effects of background strains (C57BL/6! J, C57BL/6N, C57BL/6C, 129SvEv, BALB/c), body weight, age at test, and start time of test on the results of each test, by analyzing data of more than 1200 mice (including wild type and mutant mice from 20 strains of genetically engineered mice), which had been tested in our laboratory. Also, we conducted factor analyses of a large set of data to examine the relationship

between behavioral indices, which were obtained from our behavioral test battery. The potential implications of our findings for the improvement of the behavioral test battery will be discussed.
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Phenotyping locomotor activity using automated home cage observations.
L de Visser, R van den Bos, MJH Kas, BM Spruijt
To contribute to the refinement of behavioural phenotyping methods for inbred and mutant mice, we developed a reliable tool for observing and analyzing challenge-induced and baseline behavior in a home cage-like environment (PhenoTyperÒ, Noldus Information Technology, Wageningen, The Netherlands). This method allowed for continuous, automated observations without disrupting the animals or the recording by handling or transport. Dissecting locomotor activity in detailed components and at the same time taking into account the spatial and temporal organization might facilitate the characterization of inbred strains by refining the parameters on which strains can possibly differ. Here we present the first results of a comparative study on the locomotor phenotype of four inbred strains of mice (C57BL/6, DBA/2, C3H and 129S2/Sv) using automated home cage observations. First, strain differences in locomotor activity were dependent on the time of testing (novel vs baseline conditions) due to differences in rate of adaptation to the environment. Second, Principal component analysis (PCA) was performed to study how different aspects of locomotor activity relate to each other and what that might implicate for the motivational systems underlying the behaviour. Two major components within the domain of locomotor activity were extracted by PCA, which could be interpreted as ?general activity? (or: how active is the animal?) and ?way of moving? (or: when active, how is the animal moving through the cage?). Inbred strains could be distinguished according these components, again, depending on the time of testing. The method presented here provides a valuable contribution to the refinement of the concept of the locomotor phenotype in inbred mice. The different levels and aspects that comprise the locomotor phenotype (novelty-induced, baseline, circadian rhythmicity, detailed moving patterns, long-term development) can be studied using a single setup, while minimizing human intervention.
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Estimating wall guidance and attraction in mouse free locomotor behavior.
G Horev1, Y Benjamini2, I Golani1
Measuring the path traced by a mouse in a walled arena is a common procedure in behavioral studies. Studies of neural processes associated with navigation in the arena relate momentary behavioral measurements to electrophysiological measurements. On the other hand behavior genetics studies relate cumulative behavioral measurements to genetic factors. We show that the relations between momentary location, heading direction and speed, have a heritable component, and therefore can also be used for studying gene function. We develop a method to accurately measure momentary heading direction from path data. Then we show that heading direction is influenced by the distance from the wall, and mediates the wall?s influence on speed. With increasing distance from wall, variation in heading increases gradually and speed changes gradually. For each distance, the more parallel to the wall the mouse is, the higher its speed. These interrelationships indicate two types o! f influence the wall exerts on behavior: guidance and attraction. These influences are observed in sighted and in blind mice, indicating that vision is not the only factor here. Estimation of these influences in 5 inbred strains reveals heritable components that are replicable across laboratories. This would allow studying genes mediating the wall?s guidance and attraction.
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Patterns of Fos expression in the brain of 129X1 mice selected for high exploration vs complete withdrawal in a free exploration paradigm.

F Magara, CC Marcon, JM Petit, PJ Magistretti

Mice of the strain 129X1/SvJ have often been reported to be dull and passive in a variety of behavioral paradigms, their poor proficiency possibly caused by anxiety rather than learning or motivational deficits (e.g.: Dockstadter et al., J Neurosci. 2001). In a preliminary series of experiments performed on 90-days old 129X1 mice, we observed a considerable individual variability in the coping attitude (exploration vs withdrawal) towards novel environments, with up to 40% of the mice refraining to move in response to changes of housing or testing environment. In the attempt of generating two recombinant inbred substrains differing for their trait anxiety, (neophobia vs novelty seeking), we bred to each other mice selected for high (respectively low) latencies to enter the novel compartments in a free exploration paradigm (FEP) (Misslin & Ropartz, Behav Proc 1981). This psychogenetic selection produced so far two F2 generations largely differing in activity and exploratory patterns. Taking advantage of these large individual differences, we studied the stress response, and the expression of Fos protein in the brain of animals freely exploring the novel FEP compartments, as compared to animals who did not enter, yet did not neglect, the new compartments. Albeit preliminary, results indicate that 1) Mice of the strain 129X1 show individual variability in their exploratory attitude, that is susceptible of segregation through selective breeding; 2) neophobia and agoraphobia/state anxiety, (as assessed on the elevated plus maze) can represent independent forms of anxiety, as also recognized by recent classifications of human anxiety disorders. 3) Withdrawn mice show reduced activation of hippocampal and frontal cortical fields, suggesting the involvement of these brain regions in motivational and exploratory drive. 1Centre de Neurosciences Psychiatriques, Université de Lausanne, 1008 Prilly, Switzerland and 2Life Science Faculty, Brain & Mind Institute, EPFL, 1015 Lausanne, Switzerland.

Analysis of Open Field Test (OFT) behavior of 8 inbred mouse strains using the Wall Center Separation algorithm.

D Lipkind1, A Sakov2, N Kafkafi3, GI Elmer3, Y Benjamini2, I Golani1

The OFT is one of the most widely used behavioral paradigms in mice. It is considered as a test of anxiety, exploration and locomotor activity. The common automatically collected measures in it are distance traveled and center occupancy - two simple measures that do not capture the richness and complexity of OFT behavior. A comprehensive behavioral phenotype is crucial for the elucidation of the physiological and genetic basis of complex behaviors. We aim at expanding the analysis of OFT behavior so as to create such a phenotype. Using the Software for the Exploration of Exploration (SEE) methodology, an algorithm was developed for separating the path traced by mice in the OFT into intrinsically defined patterns of movement near the wall and in the center of the arena. These patterns were used to design new measures, which provide an articulated description of various aspects of behavior in reference to the wall, and which were shown to discriminate between two inbred strains in a replicable manner. We now apply the wall/center separation algorithm to 6 additional strains, including two wild-derived strains. A detailed comparative examination shows strain differences in strategies of center occupancy, as well as in other measures of wall/center behavior.
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Common genes influence performance in mouse models of novelty seeking.

CL Kliethermes, JC Crabbe

In human populations, individuals characterized as high novelty seekers abuse drugs at a higher rate than low novelty seekers, and some neurotransmitter receptor and transporter polymorphisms have been associated with novelty seeking. Mouse models of the complex personality trait of novelty seeking allow a dissection of the genetic factors that influence the trait, and may aid in the development of therapies that will reduce the incidence of drug abuse. In order to determine if common genes influence performance in different models of novelty

seeking, we compared 14 inbred and 1 F1 hybrid mouse strains on five tests that can be putatively labeled as novelty seeking tasks, and correlated strain means from each of the tasks. These tasks included locomotion in a novel environment, head dipping, head dipping for objects, preference for a novel environment, and spontaneous alternation. Head dipping and preference for a novel environment were highly correlated with locomotion in a novel environment and modestly genetically correlated with each other; while head dipping for objects and spontaneous alternation were largely unrelated to any of the other tasks. These results imply that locomotor activity has a measure-dependent influence on task performance, and that some seemingly disparate models of novelty seeking share common genetic influences.
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Finding snail's neuropeptides in other invertebrates: a physiological study.
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Previously, a novel gene named Helix Command Specific 2 (HCS2) has been isolated from single molluscan neurons involved in control of withdrawal behavior. The predicted amino acid sequence of the HCS2 protein contains at the N-terminus a hydrophobic leader sequence followed by four putative neuropeptides, and in the C-terminal part a sequence matching the consensus motif of the EF-hand family of the Ca2+-binding proteins. All four predicted neuropeptides bear a C-terminal signature sequence Tyr-Pro-Arg-X, and three of them are likely to be amidated. These peptides called CNPs (Command Neuron Peptides) were synthesized and polyclonal antibodies were raised.
With aid of these antibodies we have found CNPs homologs in nervous system of different invertebrates: in various families of Mollusca, in several representative species of Insecta and Annelida. Using the immunohistochemistry technique we delineated a map of the CNPs-immunoreactive neurons in physiologically traceable organism, Hirudo medicinalis. At this map are described some well-known neurons (HE, Leydig), and were providing intracellular recordings for several unknown cells. Also, we tested intracellularly the neural responses to peptide CNP4 application. The role of CNPs-immunopositive cells in animal behavior and functions of CNPs neuropeptides family are discussed.
Institute of Higher Nervous Activity and Neurophysiology of the Russian Academy of Sciences. Supported by Russian Foundation for Basic Research, INTAS grant 01-2117.

Behavioral and genomic investigations of *Drosophila* soluble guanylyl cyclase mutants.
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A natural behavioral dimorphism in the fruit fly, *Drosophila melanogaster* is associated with genetic variation in the gene *foraging* (*for*). Some larvae, called 'rovers', have increased foraging locomotion compared to others, called 'sitters', and this difference is directly related to the activity of *for*-encoded cGMP-dependent protein kinase (PKG). Here we report that mutations in the gene *dgca1*, which encodes a soluble guanylyl cyclase (sGC) subunit, induce increases in both PKG activity and foraging locomotion. This is contrary to our original prediction that, based on the role of sGC in the synthesis of cGMP, *dgca1* mutant larvae would have deficient cGMP production and thus decreased PKG activation and reduced larval foraging locomotion. We performed DNA microarray analyses to compare transcriptional changes elicited by a *dgca1* mutation in both rover and sitter wildtype genetic backgrounds. We identified many genes that are differentially transcribed in either background, but interestingly, relatively few are affected in both backgrounds. Furthermore, several of these commonly affected genes are enhanced or suppressed in a background-dependent manner. Thus, genetic background has a critical influence on the molecular effects of this mutation. These findings will support future investigations of *Drosophila* foraging behavior.
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From behaviour to genes to cells: the foraging gene and its role in Drosophila melanogaster foraging behaviour.

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Although the inheritance of behaviour has interested people for ages, we have only begun to develop adequate techniques for detailed study of the relationships between genes and behaviour. Moreover, analyses of single genes that influence natural behavioural variation are particularly challenging due to the fact that individual differences in the performance of a behaviour may arise from the interaction of many genes each with small relative effects. Interestingly, natural variation in the foraging behaviour of the fruit fly Drosophila melanogaster is largely attributed to allelic differences within a single major gene called foraging (for). for encodes a cGMP-dependent protein kinase (PKG). The two foraging strategies,"rover" and "sitter", show differences in their foraging behaviours. Rovers move further in the presence of food than sitters. Molecular analyses have shown that rovers have more PKG transcript and higher kinase activity than sitters. By transgenically overexpressing for in sitters, we can transform a sitter into a rover. These findings suggest a role for PKG-mediated signaling in Drosophila foraging behaviour. We are currently investigating the expression pattern of for transcripts and PKG as well as manipulating PKG levels in order to further understand the mechanism of the foraging behaviour from genetic, molecular and neurobiological perspectives. Our findings show that PKG is localized in a cell-specific manner in the larval and adult brain as well as in the fat body of larvae and that some of these PKG expression patterns are disrupted by environmental manipulations such as acute food deprivation in larvae. By doing these types of experiments we hope to use the foraging behavior as a model system to better understand the role of genes in naturally occurring behavioural differences from genetic as well as cellular and neurobiological mechanism perspectives.

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***In vivo* single-cell control of gene expression and imaging of activity-dependent dendritic arbor growth.**

K Haas

Complex behaviors may arise from brain circuits created by interplay between genetically programmed patterns and external environmental influences. How does the external milieu influence the developing brain? Here we directly examine early stages of neuronal growth within the intact brains of developing *Xenopus laevis* tadpoles. Newly-differentiated neurons within the optic tectum receive direct glutamatergic innervation from retinal ganglion cells of the eye, and respond to visual stimuli. Remarkably, we can alter the rate of tectal neuron dendritic arbor growth, *in vivo*, by varying visual stimulation over short intervals. Single-neuron transfection strategies were employed to dissect the influence of glutamatergic transmission on tectal neuron dendritic arbor growth. Expression of the intracellular C-terminal domains of AMPA receptor subunits, GluR1 and GluR2, reduced AMPA receptor-mediated currents by approximately 50%, by interfering with protein-protein interactions necessary for receptor trafficking and insertion at synapses. Dendritic arbors of tectal neurons with reduced glutamatergic transmission grew abnormally due to the inability to stabilize new dendritic processes. Reduced glutamatergic transmission in single brain neurons produced deficits in short-term dendritic growth dynamics, long-term growth patterns, and interfered with the growth-promoting effects of visual stimulation.

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Control of inner optic chiasm development by glial expression of Drosophila optomotor-blind.

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Certain mutations in optomotor-blind (omb) that affect regulatory elements that lie far downstream of the promoter cause developmental defects in the optic lobes of the fly. In the adult animal, these are associated with defects in visual behaviour. Deletion of DNA lying 100 kb downstream of the promoter (optic lobe regulatory region 3, OLR3) causes the lack of a set of large field interneurons in the lobula plate. The lack of these cells appears to be the major cause of the eponymous optomotor-blind phenotype. Additional deletion of OLR2, located between +80 and +100 kb, causes additional structural defects in the optic lobes and exacerbates the optomotor defect. Among others, the structure of the inner optic chiasm (IOC) is perturbed. The IOC connects three neighbouring neuropil regions of the Drosophila optic lobes. Fibers that normally seem to project from and to the edges of the neuropil regions, in the mutant, appear to cut deeply into the lobula. The strength of the optomotor defect does not correlate with the apparent expressivity of the IOC defect (as measured by paraffin histology). This does not rule out that a basal defect in the projection pattern of fibres that project through the IOC contributes to the impaired optomotor performance. omb is the only Drosophila gene orthologous to a group of vertebrate paralogues Tbx2-5. Haploinsufficiency for human Tbx3 and Tbx5 and lack of omb cause similar defects in appendage development. Tbx5 is known to affect the retno-tectal projection. The current study aims to clarify the role of Tbx2-5-type T-box genes in visual development.

omb is expressed in many cell types of the developing optic lobes. ORL2+3 deletions cause the selective loss of omb expression from glial cells of the larval IOC. Loss of omb expression does not lead to the elimination of these cells which continue to express several tested glial markers. DNA fragments from ORL2 were tested for enhancer activity in transgenic flies. A 6.5 kb fragment (omb-C) drives expression in the affected IOC cell population. The UAS/Gal4 system was used to assess the relevance of omb expression for IOC formation. Restoring omb expression to omb deficient IOC glial cells (expression of UAS:omb under ombC-Gal4 control in an omb mutant background) rescued the IOC developmental defect. This demonstrates a necessary and sufficient role of omb in the IOC glia for IOC patterning.

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Specific anosmia to androstenone in inbred mice.

VV Voznessenskaya, CJ Wysocki

Studies of specific anosmias, extremes in olfactory perception, can provide insights into olfactory function, genetic regulation of olfactory phenotypes and gene/environment interactions. Using rodents as model systems, we have determined that inbred strains of mice provide an excellent genetic model of variation in sensitivity to odorants. Indeed, one is available for sensitivity to the mammalian pheromone androstenone(AND). It foundations reside in inbred CBA/J (AND-sensitive) and NZB/B1NJ (AND-insensitive) mice; however, using a behavioral endpoint, these mice could be sensitized to AND regardless of their initial level of sensitivity to the compound. CBA/J and NZB/B1NJ mice and offspring from specific matings were examined in two behavioral tests for olfactory sensitivity to AND. Thresholds to AND were estimated in a quinine aversion Y-maze test and buried cookie test. Differences in sensitivity to AND between CBA and NZB mice were estimated to be at least 2000-fold. Analysis of the results of AND sensitivity tests of the segregating F2 generation indicated probable involvement of X and Y chromosomes. Following behavioral testing genomic DNA was extracted. 98 microsatellite DNA markers were used for genome screening. Data obtained were subjected to an analysis by the software Mapmaker, which generated linkage maps (with MAP) of the markers, which conformed with published data, and generated LOD scores (with QTL) between markers and androstenone sensitivity. The two X-linked markers that were included in the genome scan did not detect any evidence for linkage. Data analysis revealed a peak LOD score between two markers on Chr 13. Adding an additional marker confirmed the linkage peak.

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The glucocorticoid antagonist Mifepristone modifies steroid signaling and coping styles in mice.

S Dalm, E Engst, ER de Kloet, MS Oitzl

Hypercortisolemia is a major characteristic of patients suffering from severe mood disorders. Interestingly, a 7-days oral treatment with a high dose of the antiglucocorticoid mifepristone (RU38486) attenuated psychotic symptoms, improved mood and cognitive performance and normalized cortisol secretion. Mifepristone is expected to modify endocrine and behavioural responses via altered steroid signaling. Male C57BL/6J mice (3-months-old) received one high dose of mifepristone (RU; 200 mg/kg via oats) either once (1x) or on seven days (7x). Control mice received vehicle-treated oats. An immediate effect of GR-antagonism is high circulating corticosterone for at least 8 hrs. However, 24-hrs later, basal corticosterone concentrations were significantly lower than in controls. Exposure to a circular hole board (5 min □novelty stress□), strongly augmented corticosterone secretion in 1xRU mice, while 7xRU were less responsive than controls. Novelty-induced ACTH remained low in the RU-treated groups. General locomotor activity was increased after 1xRU, whereas both RU-groups increased the latency to leave the center, dipping the head into the holes, and the number of holes visited using a serial pattern; 7xRU decreased the time spent in the rim area of the board. One week after cessation of 7xRU, hormonal responses and behaviour were still different. Compared to controls, mineralocorticoid receptor (MR) mRNA expression was reduced by ~20% in all hippocampal subfields after 1xRU, similar in 7x RU and again lower one week after 7xRU. Antiglucocorticoid administration has massive immediate and long-lasting effects in healthy mice. Neuroendocrine, cognitive and emotional responses are distinctly modulated, indicating an altered perception of the environment. The pulsatile blockade / activation of MR and GR most likely underlies the augmented circadian amplitude of corticosterone, leading to shifts in corticosteroid receptor balance and response patterns. The next step will be to verify therapeutic effects of mifepristone in an animal model with a dysregulated stress system.

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P66Shc knockout mice: a behavioural characterisation.

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Targeted mutation of the p66Shc gene has been shown to increases both resistance to oxidative stress and life span in a 129 Sv/Ev mouse strain. Aim of the present study was to characterise the above mentioned mutation from a behavioural point of view. To this end we compared the phenotype of p66Shc-/- (KO) to that of p66Shc+/+ wild type (WT) controls in a battery of behavioural tests at three different ages. Our results indicate that age-dependent differences present in WT animals were less pronounced in the KO phenotype, particularly in the open field and in the hot plate tests which revealed an increase in pain threshold. Data from the plus maze give an overall indication that KO mice were broadly characterised by a less anxious phenotype. In the Morris water maze, young subjects in the KO group showed a better retention when compared to all others. Studies aimed to correlate the differences seen in the emotional behaviour of these mice with selected changes in neurochemical parameters are currently in progress. In addition, the resistance to oxidative stress in the central nervous system is under investigation.

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Mice overexpressing the 5-HT1A receptor in cortex and CA3 region display different hypothermic and motor response to 8-OH-DPAT.

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Here we present the phenotyping data of mice that overexpress the 5-HT1A receptor in the outer cortical layers (I-III) and CA3 region of the hippocampus. I. We evaluated baseline behaviour and physiological functions. Contents of 5-HT and 5-HIAA were measured in selected brain areas; baseline motor activity and anxiety-related behaviour were tested in the open field and on the elevated plus maze, and additionally core body temperature was recorded. II. The response to 8-OH-DPAT (0.1-2.5 mg/kg) concerning activity, anxiety-related behaviour and body temperature was investigated. Moreover, the appearance and characteristics of the 5-HT syndrome were observed. I. No differences in 5-HT and 5-HT turnover of mutant mice were detected. Activity of transgenic mice! was slightly decreased only in the open field test. Core body temperature was significantly lower. II. The pharmacological testing revealed high impact of 8-OH-DPAT on motor skills of transgenic mice: 0.5 mg/kg 8-OH-DPAT led to a decrease of motor activity or immobility in both behavioural tests. This response was heightened during the 5-HT syndrome investigation: nearly all transgenic mice were immobile for 30 min. The 8-OH-DPAT (0.5mg/kg) induced hypothermic response was more distinct in mutant mice (3EC vs. 1EC). To summarize, the overexpressed 5-HT1A receptors are pharmacologically active. The results indicate that postsynaptic 5-HT1A receptors might be involved in thermoregulation of mice. These genetically modified mice are a promising model to further investigate the role of 5-HT1A receptors for motor abilities and the adjustment of body temperature and to study neurobiology of the serotonergic syndrome.

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Dose response of Nortriptyline and Escitalopram using the hole board and the forced swim test in four inbred mouse strains.

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In order to gain a greater inside into the interaction of genetics and pharmacology in depressive disorders a project called GENDEP has been started. The GENDEP approach is intended to connect results of behaviour, transcriptomics and proteomics on human, rodent and in vitro studies. In mouse, we are undertaking a study of genotype - environment interaction in behavioural and gene - expression responses to antidepressants. This will involve male and female mice of four inbred strains: C57BL/6J, DBA/2J, 129SvemJ and FVB/NH. As a pilot experiment we have determined a behavioural dose response with male and female mice. The animals (n=5) were treated with three different doses of two antidepressants-Nortriptyline (5mg/kg, 10mg/kg and 20mg/kg) and Escitalopram (4mg/kg, 8mg/kg, 12mg/kg). After a single dose of one drug the animals were observed in the hole board and the forced swim test. The results showed dose-dependent strain and sex related differences. Altogether the data suggested across the strains and sex that the most effective dose might be 5 mg/kg of Nortriptyline and 4 mg/kg Escitalopram.

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Maternal separation combined with isolate housing produce anxiolytic effects in C57BL/6J and DBA/2J mice.

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We recently investigated the roles of genotype and mild psychological stressors on the development of anxious phenotypes. C57BL/6J (B6) and DBA/2J (D2) mice were used to examine the effects of maternal separation and isolate housing on anxiety-like behaviors in the zero-maze. At postnatal day 12(PND 12), litters were either separated from their mother for thirty minutes per day until weaning or left undisturbed except for routine animal husbandry. On PND 21, animals were weaned and either group housed (2-5 same sex animals per cage) or isolate housed. Animals were tested in the elevated zero- maze on PND 60. We observed a significant effect of treatment on time spent in the open quadrants. Animals that underwent maternal

separation followed by isolate housing spent a larger percentage of time in the open quadrants of the zero-maze than did animals that were only maternally separated, only isolate housed, or non-maternally separated group housed. Analysis also revealed that B6 males and females spent significantly more time in the open than D2 mice. Additionally D2 males spent significantly more time in the open than D2 females. There was no significant effect of treatment on activity in the closed quadrants of the maze. B6 mice were generally more active than D2 mice. B6 females were more active than B6 males, whereas D2 females were less active than D2 males. These results concur with previous findings that B6 mice demonstrate lower levels of anxiety-related behavior than do D2 mice. The anxiolytic effect of maternal separation in combination with isolate housing may be due to the development of habituation to handling and the development of adaptive responses to these early stressors. Importantly, these findings are in agreement with others that suggest that early life experiences have a significant role in the development of adult behaviors.
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Susceptibility to activity based anorexia (ABA) in inbred and chromosome substitution strains (CSS) of mice.
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We determined trait differences in three inbred mice strains (A/J, C57BL/6J and DBA/2J) using the ABA model. These strains form the genetic background for CSS and recombinant inbred (RI) panels. These strains exhibited behavioural differences in their adaptation to food restriction. While C57BL/6J strain reduces behavioural activity during food restriction, A/J and DBA/2J mice do not show similar adaptation. The three strains have similar levels of food intake under baseline conditions, but during food restriction DBA/2J and A/J mice consume less food than the C57BL/6J strain. Finally, in contrast to A/J and DBA/2J strains, C57BL/6J mice exhibit food anticipation during food restriction. We also have tested the CSS panel in the ABA model. The CSS panel consists of 22 mouse strains each of which carries a single chromosome substituted from a donor strain (A/J) on to a common host background (C57BL/6J). Out of 22 strains, we have tested 10 strains. We have found that certain chromosomes contribute to the increased activity and suppressed food intake during food restriction. We are now in the process of testing remaining strains of the panel in the ABA model. We are aiming to perform physiological and genetic characterisation for the selected strains and perform QTL mapping for the selected chromosomes.
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Early changes in neuromotor behaviour in DREAM transgenic mice.
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Changes in gene expression underlie short-term to long-term adaptation processes within the CNS in response to changes in nuclear calcium concentration. We have characterized the DREAM protein, the first known calcium sensor with intrinsic transcriptional repressor activity that is directly regulated by changes in the concentration of Ca2+ ions (Nature 398:80-84, 1999). Expression of DREAM mRNA and protein has been observed in the CNS early during embryonic life and maximum levels are already present at birth. To understand the role of DREAM in neuronal development we have analyzed the early neuromotor behavior in wild-type mice and compared to transgenic mice over expressing dominant negative mutants of DREAM in neurons by the use of the neuron-specific CaMKII_ promoter. Embryonic expression of the dominant negative mutant in various telencephalic areas of the CNS in transgenic line 26 is correlated with an increased pivoting behavior 7 and 10 days after birth without change in the total walking activity test. Importantly, L-26 transgenic mice showed a reduced latency in the homing test at day 10 after birth supporting the notion of an accelerated neuromotor development in these transgenic animals. Thus, disruption of Ca2+/DREAM-mediated DRE- and CRE-dependent transcription during embryonic life has subtle effects in specific hallmarks of the neuromotor behavior which are not associated to generalized changes in perinatal behavior.
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β-carboline-induced seizure susceptibility and spontaneous SWDs observed in the absence-seizures are genetically correlated.

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The β-carboline compounds are inverse agonists of the benzodiazepine sites of the GABAA receptor and are potent convulsants. At lower doses, they induce behavioural arrests and spike-and-wave-discharges (SWDs) reminiscent of absence-seizures. Three independent works done in our laboratory, presented hereafter, suggest that the β-carboline-induced seizure responses and the absence-seizures are genetically correlated. 1) Two strains of mice, derived from a bidirectional selection for their seizuring response after an injection of the β-carboline methyl-beta-carboline-3-carboxylate (β-CCM), were characterized in our laboratory. The susceptible one, BS/Orl, exhibits spontaneous bilateral and synchronous SWDs upon cortical EEG, which are suppressed by anti-absence drugs. The resistant strain, BR/Orl, does not exhibit any SWDs. 2) The pseudo-replicated line to BS/Orl issued from BS/Orl.BR/Orl F2, named BS2/Orl, displays equivalent spontaneous SWDs. 3) Using the linkage-testing-strain C3XtEso, we have shown that the Bis2 gene, initially identified for its involvement in β-CCM-induced-seizures is also involved in the spontaneous SWDs control and /or genesis. Altogether, these results are converging evidence suggesting that β-CCM-susceptibility and absence-epilepsy share a common genetic inheritance.

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Systems biology approach to the brain: phenomic analysis of morphology and development of the dentate gyrus and hippocampus in inbred mouse strains.

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In the post-genomic era an important problem will be identifying the relationships between genes and brain function and anatomy. We have taken the first steps towards this goal by developing a"moderate throughput" method for assessing the affects of genetic differences on the size, shape and appearance of specific structures in the brain of the mouse. We examined and measured a large variety of architectural features of the dentate gyrus and hippocampus of 6 inbred strains (Jackson Labs, Bar Harbor, ME): A/J, BALB/cByJ, C3H/HeJ, C57BL/6J, DBA/2J, and 129X1/SvJ. All mice were males 60 ±3 d old. All measurements were made using a "landmark"based system on horizontal sections on a single level passing through the posterior commissure. A total of 36 measured or derived parameters were then analyzed with principal component analysis (SAS) which revealed that 5 underlying (and independent) "factors" accounted for most of the interstrain variation. The first 3 factors, which together account for >83% of the variance, are highly correlated with the lengths of the infrapyramidal granule cell layer, the suprapyramidal granule cell layer, and the intrusion of CA3 into the hilus, respectively. A maximum likelihood analysis (Phylip) using the 36 measurements as continuous traits shows that, from the perspective of hippocampus and dentate gyrus morphology, all 6 of these strains are statistically different from one another and that the most different strains are C3H vs BALB/c (1.98 arbitrary units apart), and the least different are A vs C57 (0.54 arbitrary units apart). In principle, the 5 "factors" detected by the principal component analysis represent at least 5 different genes acting at some time during development, but this interpretation does not consider that each of these phenotypes may represent the interaction of multiple genes. The addition of more inbred strains and recombinant inbred to this analysis will provide a basis for the mapping and future identification of the genes involved and their specific developmental functions. We are also adding microarray and behavioral data to the analysis to connect development, anatomy and function.

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Genetic mapping for ethanol-related behaviors in the LXS panel of recombinant inbred strains from ILS and ISS.

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This is the first report of QTL mapping for ethanol-related behaviors in the new recombinant inbred (RI) series (LXS) derived from Inbred Long Sleep (ILS) and Inbred Short Sleep (ISS) strains. The HS stock, from which ILS and ISS were selected, resulted from an 8-way cross of common inbred strains, producing a mosaic of their genomes in the derivative strains. This RI panel has 75 strains extant, and is the largest fully inbred mouse RI panel in existence. The genetic map consists of 330 microsatellite markers supplemented with over 15,000 SNPs. We mapped QTLs for a variety of alcohol-related traits, including low-dose activation (LDA), loss of righting (LORR), BEC at awakening, and hypothermia (HYPO). Some of these QTLs replicate regions reported previously, others are novel. When confidence intervals of these QTLs overlapped previously reported QTLs, we combined p-values using Fisher's exact test. Some of the combined p-values are highly significant, providing strong support for real QTLs. Data and mice are available to the mouse research community. This work was supported by NIH: (RO1AA11984 (TJ); KO1AA00195 (TJ); P50A13755 (RW)), the Ellison Foundation (TJ), and funds from the University of Colorado. Genotyping costs were supported in part by P20MH 62009 (RW) and the INIA Genotyping Core.

Genetic contribution to antisocial traits and alcoholism in Native Americans.

CL Ehlers, DA Gilder, KC Wilhelmsen

Gene and gene X environment studies of complex diseases, such as alcohol dependence and antisocial personality disorder, have advantages when conducted in well-defined populations such as Indian tribes. Often such populations are more environmentally and genetically homogeneous, they are geographically more restricted, and large extended pedigrees are more common than in the general population. This study conducted genetic linkage analyses to conduct disorder and ASPD phenotypes, and explored their interaction with alcohol dependence in Mission Indians (n=467). Alcohol dependence and antisocial diagnoses were made using the SSAGA. Variance component estimate methods were used to calculate LOD scores using SOLAR. Alcoholism was found to be significantly comorbid with ASPD in this population. Several measures of antisocial behavior were found to aggregate in Mission Indian families suggesting moderate heritability (Hr=.33-.5). Suggestive LOD scores (2-3) were detected on chromosome 2, 10 and 13 for ASPD. Data from this study provide support for the linkage results from COGA for conduct disorder for chromosome 2. Bivariate analysis with ASPD and Antisocial Problems enhance the LOD score for two putative loci previously identified for alcohol withdrawal on chromosome 15. Additional families have been ascertained for this study that will allow for confirmation. Further studies of candidate genes in the locations studied may provide the framework to study the relationship between ASPD/conduct and alcohol and drug dependence.

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