Title

Issues related to symptomatic and disease-modifying treatments affecting cognitive and neuropsychiatric comorbidities of epilepsy.

Journal

Epilepsia, 54 Suppl 4(0 4)

ISSN

0013-9580

Authors

Brooks-Kayal, Amy R
Bath, Kevin G
Berg, Anne T
et al.

Publication Date

2013-08-01

DOI

10.1111/epi.12298

Peer reviewed

Published in final edited form as:
Epilepsia. 2013 August ; 54(0 4): 44–60. doi:10.1111/epi.12298.

Issues related to symptomatic and disease-modifying treatments affecting cognitive and neuropsychiatric comorbidities of epilepsy

Amy R. Brooks-Kayal[*], **Kevin G. Bath**[†], **Anne T. Berg**[‡], **Aristea S. Galanopoulou**[§], **Gregory L. Holmes**[¶], **Frances E. Jensen**[**], **Andres M. Kanner**[††], **Terence J. O'Brien**[‡‡], **Vicky H. Whittemore**[§§], **Melodie R. Winawer**[¶¶], **Manisha Patel**[***], and **Helen E. Scharfman**[†††]

[*]Departments of Pediatrics, Neurology and Pharmaceutical Sciences, University of Colorado Schools of Medicine and Pharmacy, Children's Hospital Colorado, Aurora, Colorado, U.S.A.

[†]Department of Neuroscience, Brown University, Providence, Rhode Island, U.S.A.

[‡]Ann & Robert H Lurie Children's Hospital of Chicago, Chicago, Illinois, U.S.A.

[§]Saul R. Korey Department of Neurology, Dominick P. Purpura Department of Neuroscience, Laboratory of Developmental Epilepsy, Albert Einstein College of Medicine, Bronx, New York, U.S.A.

[¶]Departments of Neurology and Pediatrics, Geisel School of Medicine at Dartmouth, Hanover, New Hampshire, U.S.A.

[**]Department of Neurology, University of Pennsylvania, Perlman School of Medicine, Philadelphia, Pennsylvania, U.S.A.

[††]Department of Neurology, University of Miami, Miller School of Medicine, Miami, Florida, U.S.A.

[‡‡]Department of Medicine (Royal Melbourne Hospital), The Melbourne Brain Centre, University of Melbourne, Parkville, Victoria, Australia

[§§]National Institute of Neurological Disorders and Stroke, National Institutes of Health, Bethesda, Maryland, U.S.A.

[¶¶]Department of Neurology, The G.H. Sergievsky Center, Columbia University, Manhattan, New York, U.S.A.

[***]Department of Pharmaceutical Sciences, University of Colorado Skaggs School of Pharmacy and Pharmaceutical Sciences, Aurora, Colorado, U.S.A.

[†††]Departments of Psychiatry, Physiology & Neuroscience, The Nathan Kline Institute of Psychiatric Research, New York University Langone Medical Center, New York, New York, U.S.A

Address correspondence to Amy Brooks-Kayal, Chief and Ponzio Family Chair in Pediatric Neurology, 13123 East 16th Avenue, Box B155, Aurora, CO 80045, U.S.A. Amy.brooks-kayal@childrens-colorado.org.

The authors confirm that they have read the Journal's position on issues involved in ethical publication and affirm that this report is consistent with those guidelines.

Disclosures

Dr. Berg has received speaker honoraria and travel support from BIAL and MUSC, and serves on advisory boards for CURE and Eisai. Dr. Galanopoulou has received speaker honorarium from Novartis. Dr. Kanner has served as member of a data safety board for Vertex. Dr. O'Brien has received research grant support from the NHMRC (Australia), the Royal Melbourne Hospital Neuroscience Foundation, Sanofi-Aventis, UCB, SciGen, GlaxoSmithKline, Novartis and Janssen-Cilag, and speaker honorarium from Sanofi-Aventis, UCB, SciGen, GlaxoSmithKline, and Janssen-Cilag. None of the other authors have any conflicts of interest to report related to this manuscript.

Summary

Many symptoms of neurologic or psychiatric illness—such as cognitive impairment, depression, anxiety, attention deficits, and migraine—occur more frequently in people with epilepsy than in the general population. These diverse comorbidities present an underappreciated problem for people with epilepsy and their caregivers because they decrease quality of life, complicate treatment, and increase mortality. In fact, it has been suggested that comorbidities can have a greater effect on quality of life in people with epilepsy than the seizures themselves. There is increasing recognition of the frequency and impact of cognitive and behavioral comorbidities of epilepsy, highlighted in the 2012 Institute of Medicine report on epilepsy. Comorbidities have also been acknowledged, as a National Institutes of Health (NIH) Benchmark area for research in epilepsy. However, relatively little progress has been made in developing new therapies directed specifically at comorbidities. On the other hand, there have been many advances in understanding underlying mechanisms. These advances have made it possible to identify novel targets for therapy and prevention. As part of the International League Against Epilepsy/American Epilepsy Society workshop on preclinical therapy development for epilepsy, our working group considered the current state of understanding related to terminology, models, and strategies for therapy development for the comorbidities of epilepsy. Herein we summarize our findings and suggest ways to accelerate development of new therapies. We also consider important issues to improve research including those related to methodology, nonpharmacologic therapies, biomarkers, and infrastructure.

Keywords

Epilepsy; Comorbidity; Animal models; Biomarkers; Therapy

Over the past decade, there has been increasing recognition that epilepsy is a spectrum disorder that is defined by spontaneous recurrent seizures, but for many people, epilepsy also includes a number of comorbid conditions that substantially affect their health and quality of life. These include cognitive impairment, depression, anxiety, attention deficit, autism, suicidality, psychosis, and migraine. Health concerns related to cardiac function, obesity, and sleep are also common. These diverse comorbidities present a tremendous problem for people with epilepsy and their caregivers, decreasing the quality of life, complicating treatment, and increasing mortality. The 2012 Institute of Medicine (IOM) Report, entitled “Epilepsy Across the Spectrum,” emphasized this point: “Individuals with epilepsy are at risk not only for seizures, but also for a myriad of comorbid health conditions…. Often the comorbidities that accompany epilepsy outweigh the burden of the seizures themselves” (Gilliam et al., 2003; Institutes of Medicine Committee on Public Health Dimensions of the Epilepsies, 2012). Despite the increasing recognition of the frequency and impact of cognitive and neuropsychiatric comorbidities of epilepsy, highlighted in the 2012 IOM report and the National Institutes of Health (NIH) Benchmarks, relatively little progress has been made in developing new therapies directed specifically at comorbidities.

However, there have been considerable advances in understanding the underlying mechanisms that contribute to comorbidities in epilepsy. For example, there is increasing evidence for shared bidirectional pathophysiology between depression and epilepsy (Kanner, 2011; Adelow et al., 2012; Hesdorffer et al., 2012). Studies in animal models have demonstrated shared mechanisms and genetic predispositions between seizure susceptibility and behavioral phenotype (Groticke et al., 2007; Tabb et al., 2007; Groticke et al., 2008; Kim et al., 2009; Mazarati et al., 2010; Pineda et al., 2010; Sarkisova & van Luijtelaar, 2011; Takechi et al., 2011). In addition, there have been advances in the identification of

potential targets for therapy and prevention (Johnson & Shorvon, 2011; Mula, 2012). As part of the International League Against Epilepsy/American Society for Epilepsy (ILAE/AES) workshop on comorbidities, our working group addressed the current state of understanding of the comorbidities. Herein we define terminology related to studies of comorbidities, methods, and we suggest ways to accelerate development of new therapies. We also consider issues specific to preclinical research including those related to study design, evaluation of nonpharmacologic therapies, biomarkers, and infrastructure.

Terminology

Basic terms

The following terms are commonly used when discussing comorbidities, but are often used with different meaning. Therefore, we suggest definitions for future use that can potentially unify the field and avoid confusion.

Comorbidity—A condition that occurs in association with another (i.e., epilepsy) at frequencies that are significantly greater than those observed in the appropriate control group. A comorbidity may be a cause of epilepsy, a consequence of epilepsy, or a separate condition that is associated with epilepsy because there is a common cause for the epilepsy and the comorbidity. It is important to note that in the future, some of these comorbidities could be shown to result from a spurious association, in which case they would not be true comorbidities.

Cause or causal association—A cause is a factor that contributes to the occurrence of a disease or other outcome. Manipulation of the cause leads to a corresponding change in that outcome. The link between cause and effect may be direct or indirect. The understanding of the connection between cause and effect may occur at many levels, from societal to molecular. The appropriateness of each level depends on the purpose of the investigation and the type of explanation sought or intervention planned. Causes may be (1) necessary (i.e., the presence of the "cause" is mandatory for the effect), (2) sufficient (i.e., the cause is all that is required to produce the effect), or (3) contributory (i.e., associated with an increased occurrence of the effect but typically neither necessary nor sufficient). A cause may be necessary but not sufficient, or sufficient but not necessary.

Causal pathway—This is the series of events and/or mechanisms between an initiating cause and an effect. The more complex the path from the cause to the final outcome, the less direct and weaker is the association between the initiating event and the ultimate effect.

Shared causation—This occurs when two outcomes are independent but a consequence of the same causal factor.

Confounding—Confounding can occur when two factors (A and B) are statistically related, because they are both associated with a third variable (C). To be confounding in the association between A and B, C must be a cause of A and be correlated with B but not be part of the causal pathway between A and B. C may be a cause of B or just associated with B.

Types of comorbidities

Based on the definitions described earlier, the following relationships between comorbid conditions can be defined:

Essential comorbidity—This phrase refers to two conditions that share an underlying causal factor. For example, both cerebral palsy and epilepsy may be consequences of an early brain insult. Developmental/genetic encephalopathy (described further below) conceptualizes intellectual disability and seizures as being essential comorbidities that are caused by the same underlying condition.

> *Developmental/genetic encephalopathy:* Many factors that cause intellectual disability and related behavioral consequences are associated with epilepsy. It is critical to distinguish between cognitive effects that are due to the underlying cause versus those due to seizures. The concept of a developmental-genetic encephalopathy is that the cause itself produces comorbidities. Tuberous sclerosis complex is an example in which genetic mutations that cause the disorder affect fundamental neurobiologic phenomena (mammalian target of rapamycin [mTOR] signaling) that can lead to cognitive deficits—independent of seizures (Ehninger et al., 2008; van Eeghen et al., 2012a,b).

Secondary comorbidity—The term "secondary comorbidity" is used when one condition is involved directly in producing a second condition. The second condition is the secondary comorbidity. For example, epileptic encephalopathy (defined below) conceptualizes intellectual disability and cognitive impairments as secondary comorbidities of seizures. The implication is that effective intervention for one (seizures) may lessen frequency or severity of the other (intellectual disability).

> *Epileptic encephalopathy:* This term refers to a process defined in the 2010 Classification and Terminology report of the ILAE when "the epileptic activity itself may contribute to severe cognitive and behavioral impairments above and beyond what might be expected from the underlying pathology alone (e.g., cortical malformation), and…can worsen over time." (Berg et al., 2010).

"Iatrogenic" comorbidity—This term is used when the treatment of one condition leads to or exacerbates another condition. For example, many drugs that are used to treat epilepsy can induce cognitive and behavioral impairments.

"Situational or contextual" comorbidity—These terms refer to social–environmental factors that, as a consequence of one condition, may have an impact on an individual and influence the occurrence of another condition. For example, uncontrolled seizures may lead to the loss of driving privileges, which may in turn result in unemployment, social isolation, and depression.

Complex/interacting comorbidities—These comorbidities involve multiple mechanisms that may be interdependent. For example, cognitive impairment that is a comorbidity of epilepsy may be produced by seizures or by medications to treat seizures. Furthermore, comorbidities may have different relationships to one another. In some instances, they may be mutually reinforcing. For example, sleep disorders are a common comorbidity of epilepsy. Sleep disorders are comorbid with depression; they may cause depression or be exacerbated by depression.

Animal Models to Study Comorbidities of Epilepsy

Animal models provide an opportunity to investigate the pathophysiology of comorbidities of epilepsy, and to explore the temporal relationship between the comorbidity and epilepsy. The temporal relationship is important because some comorbidities begin prior to the onset of epilepsy, whereas others arise after epilepsy is established. Furthermore, animal models

provide an opportunity to address the effects of therapeutic interventions (Blumenfeld et al., 2008; Russo et al., 2011).

Consistent with the clinical evidence that many types of epilepsy are associated with an increased incidence of comorbidities, cognitive and behavioral impairments have been reported for diverse animal models of epilepsy. Commonly used rat models of acquired epilepsy, which use kindling, status epilepticus, or traumatic brain injury to induce epilepsy, are accompanied by behavioral abnormalities, including increased anxiety- and depression-like phenotypes (Kalynchuk, 2000; Milman et al., 2005; Mazarati et al., 2007; Jones et al., 2008a) and cognitive deficits (Hamm et al., 1993; Hannesson & Corcoran, 2000; Detour et al., 2005; Kemppainen et al., 2006; Jessberger et al., 2007; Chauviere et al., 2009). The two most commonly studied rat models of genetic generalized epilepsy (GGE) with absence seizures, Genetic Absence Epilepsy Rats from Strasbourg (GAERS) and WAG/Rij rats, also display cognitive impairments and behavioral abnormalities, and these phenotypes have some similarity to psychiatric symptoms of people with GGE (Ott et al., 2003; Sarkisova et al., 2003; Jones et al., 2008a; Sarkisova & van Luijtelaar, 2011), as well as psychosis (Jones et al., 2010). Intriguingly, the behavioral phenotypes of GAERS are manifested before the onset of the seizures, suggesting that the behavioral deficits are not a secondary effect of the epilepsy (Jones et al., 2008a). Many of the behavioral comorbidities in common rodent models of epilepsy are summarized in Table 1.

In addition, there are animal models (described below) that are potentially useful to study comorbidities in epilepsy but are not common in epilepsy research. These “lesser known” models are being used to study psychiatric illness. Some of the seizures in these animals are spontaneous and repetitive, criteria that define epilepsy. Some of the methods to induce epilepsy, such as early life injury, are relevant to human epilepsy (e.g., temporal lobe epilepsy; TLE). Therefore, there is a great opportunity to use these models to study comorbidities in epilepsy.

The definition of comorbidity often depends on the perspective of the investigator: The investigator studying schizophrenia may consider the seizures in the neonatal ventral hypothalamic lesion (NVHL) model (Lillrank et al., 1995; Lipska et al., 1995; Beerpoot et al., 1996), a comorbidity, whereas the investigator interested in epilepsy may consider this animal model to be an excellent animal model of epilepsy where schizophrenia-like symptoms emerge as a comorbidity. More research is needed to clarify which symptoms arise first, the seizures or the psychiatric symptoms, to help address the problem. Until now, the video-electroencephalography (EEG) studies needed for these assessments have not been feasible. Conducting the video-EEG studies in the NVHL model is an example of a way that an NIH benchmark—to create/validate more models of epilepsy with comorbidities—could be addressed. The NVHL model is interesting for other reasons also: One example is that experiments have recently shown that psychiatric symptoms may be modifiable by behavioral interventions during puberty (Lee et al., 2012). Epilepsy researchers can use such studies to provide starting points for potential behavioral interventions that might reduce comorbidities in other animal models of epilepsy, and eventually in humans.

Many investigators are currently interested in shared causes of epilepsy and comorbidities. Supporting the case for a shared causation between the epilepsy and psychiatric comorbidities, rat strains that have an increased predisposition to developing epilepsy following a brain insult (but are not epileptic initially), such as the FAST rats (a strain where kindling occurs rapidly) and genetically epilepsy prone rats (GEPRs), show a range of behavioral phenotypes relevant to the psychiatric comorbidities of patients with epilepsy (Anisman & McIntyre, 2002; McIntyre et al., 2004; Jobe & Weber, 2005; Gilby et al., Runke et al., 2011). In mouse models of Alzheimer’s disease (AD), which often exhibit

recurrent seizures and therefore have epilepsy (Palop et al., 2007; Minkeviciene et al., 2009; Scharfman, 2012b), cognitive impairments that accompany seizures may be due to similar mechanisms as the seizures. For example, it has been suggested that one of the mechanisms underlying the cognitive impairments and the seizures are dysfunctional sodium channels, specifically NaV1.1 (Verret et al., 2012). Notably, mutations in NaV1.1 are considered to be the cause of two genetic epilepsy syndromes, genetic epilepsy with febrile seizures plus (GEFS+), and severe myoclonic epilepsy of infancy (SMEI), which is also known as Dravet syndrome, and is accompanied by cognitive deficits (Catterall et al., 2010; Scharfman, 2012a).

Guidelines for Preclinical Studies of Comorbidities

General considerations

The design of any preclinical study that addresses treatment of comorbidities should be clearly outlined. This includes description of the comorbidity (i.e., specific symptom vs. a defined syndrome or disease), the type of comorbidity (essential, secondary, iatrogenic, situational/contextual, or complex/interacting), and the type of the study (e.g., proof of concept, safety/tolerability, biomarker validation).

Several recent publications have outlined recommendations for the design of such studies and guidelines for reporting, to ensure rigor and transparency (Kilkenny et al., 2010; Rigor in Science Workgroup N, 2011; Galanopoulou et al., 2012; Landis et al., 2012). These include: justification of species, strain and experimental model (s), description of testing procedures, justification of drug chosen for study and its formulation, description of drug delivery, description of power analysis and selection of appropriate sample sizes and statistics, randomization procedures, description of inclusion and exclusion criteria, and reporting of missing data. Blinding, although desirable, may be difficult for studies that aim to treat behavioral comorbidities and needs to be stated as a limitation. Unbiased reporting and discussion of all results (negative or positive), discussion of studies supporting or disputing the findings, and disclosure of conflicts of interests are all important. Establishing clinical relevance of preclinical studies, and identifying appropriate target patient populations for studies of epilepsy comorbidity treatment entail some intricacies in design and interpretation that merit special attention, and are discussed below.

Targeting symptoms versus clinically defined syndromes

The Diagnostic and Statistical Manual of Mental Disorders, 4th edition, Text Revision (DSM-IV-TR; American Psychiatric Association, 2000), provides diagnostic criteria for the majority of psychiatric disorders, based on a consensus opinion reached by a group of experts. However, as our understanding of the clinical semiology of depressive disorders (and other psychiatric comorbidities) has advanced, it has become clear that the proposed classification is incomplete (Kanner et al., 2010). Indeed, in the last decade, psychiatrists have recognized the importance of subtypes of depression (Kanner, 2011).

Most preclinical anticomorbidity studies use behavioral tests that target specific behaviors at predefined time points that may not necessarily parallel clinical diagnostic criteria. These limitations could affect the ability of the preclinical studies to address the clinical disorder. Table 2 exemplifies the complexity of reaching a diagnosis of “depression” in an animal model based on the clinical diagnostic criteria. To adapt the DSM-IV-TR criteria for major depressive disorder for rodents would require longitudinal observations and developing a method—likely to be difficult—to identify the “personality” of a rodent and distinguish normal from depressed personality. In many preclinical studies of epilepsy-related comorbidities, assessment of depressive disorder is based on tests and conditions that vary from laboratory to laboratory. A list of commonly used rodent behavioral tests and what

they are used to assess are provided in Table 3. The tests are usually performed at consecutive time points that may span several weeks. In epileptic animals, where the epileptic condition may be evolving, disease progression over several weeks may make the results difficult to interpret. It is also important to note that some tests, like the saccharin preference test or the Porsolt forced swim test (Epps et al., 2012) assess only criteria A1, A2, and perhaps partially A3 of the adapted DSM criteria (Table 2). Furthermore, many of the tasks used to assess affective or cognitive functioning, such as the Porsolt forced swim test, can only be employed once because the task involves stress, which may alter behavior and lead to a different response if the task is conducted a second time. Although such tests have been successful in identifying drugs that improve depressive symptoms in normal animals, examples of pharmacologic studies in epileptic animals are rare. Additional tests such as novelty-suppressed feeding have also been useful to detect effects of selective serotonin reuptake inhibitors (SSRIs) in normal animals (David et al., 2007) but may be difficult in epileptic animals because of the potential effects of caloric restriction on seizures (Hartman et al., 2010). However, if the goal is to identify disease-modifying therapies for comorbidities, it may be important to utilize diagnostic criteria that consider the comorbidity as a disease complex or syndrome rather than defining it by individual symptoms.

An interesting and potentially informative caveat to treatment studies targeting comorbidities in epileptic animals is that drugs used in humans to treat a comorbidity may affect seizures. For example, drugs used for the treatment of depression, such as SSRIs and tricyclic antidepressants, exert antiseizure effects in several animal models of epilepsy. In a model of generalized epilepsy in which generalized tonic–clonic seizures are induced by audiogenic stimuli (the Genetically Epilepsy Prone Rat [GEPR] Jobe et al., 1973), abnormal serotonergic axon arborization has been identified, and deficient postsynaptic 5-hydroxytryptamine receptor 1A ($5HT_{1A}$) receptor density is present in the hippocampus. Increases in 5HT levels with the SSRI sertraline resulted in a dose-dependent reduction in seizure frequency, which correlated with the serotonin concentration in the thalamus (Yan et al., 1995). In addition, it has been shown that the 5-HT precursor 5-hydroxytryptamine (5-HTP) has anticonvulsant effects in GEPR rats when combined with a monoaminoxidase inhibitor (MAOI) (Jobe et al., 1999). Similarly, a role of 5HT and antiseizure effects of SSRIs has been suggested in multiple focal epilepsy models in several species (Prendiville & Gale, 1993).

For the accurate measures of the signs and symptoms of a disorder, the current approach to behavioral phenotyping may need to be significantly modified. In addition, as mentioned earlier, current approaches often fail to satisfy the requirement of measuring sustained changes in symptoms. The fact that many tasks provide a snapshot of behavior and cannot be repeated illustrates the need to use multiple assays at varied intervals (which themselves could induce affective symptoms) or to develop novel methods. Currently, new computerized approaches are being developed to automate tracking and cataloging of behaviors in the homecage (Jhuang et al., 2010; Lin et al., 2011). Such approaches are noninvasive and could provide more detailed quantification of routine behavior that may be a more sensitive indicator of behavioral symptoms of affective pathology (e.g., sleep disturbances, altered food intake, lethargy, and impaired social behaviors). These methods are in their “infancy,” and will require validation by comparison to existing methods, but could significantly enhance the ability to detect and track behavioral comorbidities.

Choice of animal model and species

The selection of species, strains, and genetic background should be based on the availability of accepted animal models with a reproducible clinically relevant phenotype of the seizure-related comorbidity. Validation that spans species, strains, or models would support the value of a treatment, but it would be associated with significant cost, resources, time, and

animal use. Interpretation of the results is also complicated in situations where there is incomplete knowledge about the pathogenic mechanisms of a comorbidity or the target relevance in the animal model that is chosen. Therefore, with the exception of iatrogenic comorbidities, for which testing in at least two species (rodent and nonrodent) may be required to meet regulatory requirements, testing many species for early preclinical studies is desired but not required.

On the other hand, validation of the efficacy of a treatment in at least two preclinical studies of the same seizure-related comorbidity, performed in independent laboratories using acceptable standards of testing, is strongly recommended before transition to clinical trials. Such studies could be from the same or different species, strains, or animal models. Incongruent results across studies may not necessarily disprove the findings, if the observed discrepancies can be justified by differences in target relevance or engagement or other identifiable differences in the study protocol or experimental conditions. Reporting conditions used for housing and breeding, genetic background, and procedures for handling animals should also be described.

Age and sex

Seizures, comorbidities, and response to treatments are all influenced by age and sex. Age and sex also influence the interactions between seizures and comorbidities. As a result, age and sex should be addressed in the experimental design of a study. When immature animals are used, age merits additional considerations because epileptogenesis may require time, and be faster or more severe at some ages than others. In addition, epilepsy is not a static condition but dynamic, even in the adult, so superimposing the changing nature of the epileptic condition on the changing nature of a rapidly developing or aging nervous system poses additional challenges. Age-matched controls are needed. Although one sex may be sufficient for a single study, or in cases where a comorbidity occurs only in one sex, testing in both sexes is desired—both for efficacy and tolerability.

One consideration that is important but rarely addressed directly is the interaction of age and sex of the species. In early neonatal life, when many developmental models of epilepsy are studied, circulating levels of 17β-estradiol are critical to normal central nervous system (CNS) development, and this is true for both males and females (McCarthy, 2010; Konkle & McCarthy, 2011). In males, a neonatal surge in testosterone occurs, and testosterone is aromatized to estradiol. Estradiol influences the development of many brain circuits at this early stage of development (McCarthy, 2010; Konkle & McCarthy, 2011). Many of these areas of the brain are affected by seizures or influence seizures, and are likely to influence social behavioral tests (and possibly other behavioral tests as well).

Interactions between seizures and comorbidities

As discussed previously, the relationship between seizures and a comorbidity may be complex, and regulated by many variables, some of which are hard to define. A comorbidity may share risk factors with epilepsy, may directly or indirectly be triggered or accentuated by seizures, their sequelae or their treatments, or may predispose a study subject to have seizures. An optimal study design would anticipate such interactions and control for them. Comparison to appropriate controls to determine the incidence (if any) and/or natural course of the comorbidity in a nonepileptic animal population matched for strain, treatments, handling, and living conditions would be useful. In many cases this detailed characterization may be beyond the scope of an individual study, but relevant discussion regarding the limitations of data would be helpful. Clarifying how treatments affect these interactions may be important in identifying the appropriate patient populations in future clinical trials.

Treatment formulation and delivery

There are three important issues to consider in relation to treatment formulation and administration: (1) the formulation, (2) handling, and (3) dose–response relationships. Regarding the choice of treatment formulation and delivery method, each has the potential to influence or be influenced by the targeted comorbidity. For instance, the use of oral formulations in subjects with reduced food intake (due to loss of appetite) caused by depression may limit drug exposure and underestimate its efficacy. Another example of the complex effects of drug formulation is the effect of drug formulations on a response to placebo, where the drug formulation may generate a placebo effect selectively in one treatment population. For example, sweet oral formulations may cause a placebo effect that is robust in rats with sucrose preference but not in rats without this preference. Handling the animals just before, during, or immediately after delivery may also generate confounding effects, especially when behavioral or cognitive endpoints are evaluated. Therefore, handling should be practiced in the same manner across treatment groups. Dose–response studies are helpful in defining the safety-efficacy profile of the treatment. Definition of the therapeutic window for treatment administration using clinically relevant time points would be most helpful in translating the findings to clinical trials.

Monitoring outcomes

Unbiased, blinded, and quantitative methods to monitor outcomes are strongly encouraged. Parallel monitoring of both seizures and comorbid conditions would be helpful to determine whether benefit (or lack thereof) is due to a change in seizures or comorbidities. Parallel assessment of efficacy as well as safety in early stages of therapy development is advised. The goals are (1) to document drug exposure, (2) to define a therapeutic window, (3) to identify health risks that would limit drug development, and (4) to determine the effects of treatment in individuals with epilepsy, because it may differ drastically from the response of the normal population. In studies of disease modification, assessment at least once after there is a drug effect and after drug cessation (“washout”) is desired. To document persistence of a drug effect, assessment at a minimum of two appropriate time points are advisable, with the duration of each evaluation and time between evaluations of sufficient duration to define the effect and document recurrence.

Statistics

In addition to the general issues of statistical analysis mentioned earlier, the variability of the targeted phenotype within the tested animal population (across animals, transiency of phenotype) should be accounted for in the power analysis and study design. Age, sex, environment, circadian rhythm, genetic background, methods for handling, testing conditions, and seizure frequency should be analyzed as covariates.

Reporting results

As mentioned previously, reporting of both negative and positive effects is essential. In addition, providing evidence for target engagement in the appropriate part of the CNS is critical in distinguishing drug failure from insufficient drug exposure or inappropriate experimental design. Definition of the targeted comorbidity and the subset of patients with epilepsy that exhibit the comorbidity (“target population”) are important. In addition to reporting the effects of treatment on seizures and comorbid conditions, the therapeutic window, nature of treatment (symptomatic vs. disease modifying), and candidate biomarkers would be useful to facilitate translation. Discussion of the barriers to progress to clinical testing would be helpful also. For example, the results of a particular study may suggest a need to consider individualized therapies rather than conventional clinical testing.

Examples of Approaches to Specific Comorbidities

Cognitive impairment

In individuals with epilepsy there is an associated high rate of cognitive difficulties that compromise educational progress and achievement during adult life (Davies et al., 2003; Berg et al., 2008). A deficiency in at least one academic area using an IQ achievement–discrepancy definition is identified in about 50% of children with epilepsy (Fastenau et al., 2008), and in between 40% and 60% of children with epilepsy using a low achievement definition (Fastenau et al., 2008). These effects are present even in those functioning at or near an average IQ (Oostrom et al., 2003, 2005). In the school setting, approximately 45% of children with epilepsy and an IQ of at least 80 require special education services, and 16% of these children repeat a year of education (Aldenkamp et al., 1990; Berg et al., 2011). There is increased academic risk in terms of both reading skills (including single word, phonologic awareness, comprehension, and speed or fluency deficits) and math skills (Bailet & Turk, 2000), and often several educational areas are affected in individuals. In addition to these specific academic deficits, children with epilepsy often show general deficits in memory and attention (Metz-Lutz et al., 1999).

The approach to treating a cognitive comorbidity has been primarily through nonpharmacologic interventions such as special education in children and cognitive rehabilitation in adults. The basis for this approach lies in the understanding that there is substantial plasticity in the CNS. Studies over the last decade have demonstrated that neuronal networks are plastic and that connectivity is remodeled throughout life by changes at synapses, as well as other mechanisms (Caroni et al., 2012).

There is evidence in animal models that extensive training (“overtraining”) may ameliorate cognitive impairments in some types of epilepsy. For example, it has been shown that rats exposed to early life seizures exhibit impairments in hippocampal-dependent tasks in adulthood. However, if the rats are overtrained they eventually perform the task as well as controls, which may be related to the ability to increase theta or gamma oscillations (Kleen et al., 2011).

In addition to over-training, environmental enrichment has been effective in reversing spatial learning and memory deficits in rodents following status epilepticus (Rutten et al., 2002). Environmental enrichment prior to status epilepticus reduced apoptosis and the risk of spontaneous seizures after status epilepticus (Young et al., 1999). However, in other animal models, effects of environmental enrichment are varied. In an animal model of atypical absence epilepsy, environmental enrichment improved olfactory recognition and spatial learning, but did not significantly reduce the number of ictal discharges (Stewart et al., 2006). In transgenic mice with a missense mutation of the sodium channel Scn2a (Nav1.2) and recurrent seizures, enriched sensorimotor experience from birth reduced hyperexcitability and histopathologic changes in the mice (Manno et al., 2011). It is important to note that the concept of environmental enrichment, although intriguing, is important to consider carefully. One reason is that most laboratory animals have little enrichment in their typical laboratory environment, which is different from an animal in its natural setting, and different from humans.

The ketogenic diet appears to have a positive effect on cognition in some studies of rodents but not all. For example, in aged rats, the ketogenic diet improved cognition (Xu et al., 2010). In contrast, the ketogenic diet did not appear to influence cognition in normal adult rats (Thio et al., 2010). Zhao et al. (Zhao et al., 2004) found that the ketogenic diet had an adverse effect on spatial learning and memory. However, in amygdala-kindled adult rats, the

ketogenic diet did not adversely affect spatial learning or exploratory behavior (Hori et al., 1997).

Depression and anxiety

Depression is a common psychiatric comorbidity in epilepsy. Many hypotheses have been proposed to explain the relationship between depression and epilepsy. For example, Koh et al. (2007) suggested that seizures lead to increased susceptibility to depression by reducing expression of serotonin receptors. This study was an excellent example of an approach to gain insight into a comorbidity using an animal model. The investigators used the kainic acid model, where status epilepticus is induced by systemic administration of kainic acid, and there are chronic spontaneous seizures that develop in the subsequent weeks. The authors induced status epilepticus and then investigated differential gene expression using microarray, reverse transcriptase polymerase chain reaction (RT-PCR) and the Porsolt forced swim test. The authors reported that the gene for one of the serotonin-receptor subtypes was reduced following status epilepticus and that the changes in the gene were paralleled by decreased mobility in the Porsolt forced swim test. Of interest, they also showed that a potential treatment was effective: There was improved behavior in rats reared in an enriched environment. Another study used exploratory behavior to address anxiety. Rats exhibited a reduction in exploration in the open field test after kainic-acid administration. However, rats that were exposed to an enriched environment were similar to controls (Koh et al., 2005). In an animal model of atypical absence epilepsy, environmental enrichment also improved performance on tasks that are used to evaluate anxiety (Stewart et al., 2006).

Biomarkers of Comorbidities

A biomarker is as an indicator that can be measured objectively and reflects a normal biologic process, the development of disease, or therapeutic intervention (Biomarkers Definitions Working Group, 2001). It should be noted that this definition distinguishes a biomarker from surrogate endpoints that specifically predict clinical outcomes. Within this definition of “biomarker,” genetic, behavioral, anatomic, or metabolic change can all serve as biomarkers, and they can reflect the severity of seizures or a comorbidity, or both.

As biomarkers for comorbidities of epilepsy are developed, it is important that they reflect (1) changes in the disease with time (e.g., progression) and (2) have predictive value for efficacy of therapeutic intervention. It is also valuable if they relate to a mechanism for the comorbidity. Because comorbidities may cause greater impairment in quality of life than seizures do, developing biomarkers of comorbidities of the epilepsies is equally important to developing biomarkers of epileptogenesis. Consideration should be given to biomarkers that reflect both the permanent impairments related to the epilepsy and transient impairments, which may be associated with ongoing seizures and/or medication. Some biomarkers may reflect impairments that contribute to epilepsy (e.g., neuronal damage or loss, synaptic reorganization, inflammation). Other biomarkers, such as interictal spikes and altered behaviors, may be related to dynamic impairments caused by seizures. In considering biomarkers that reflect comorbidities, the following classification is useful. Different types of biomarkers are listed in Table 4, with the acknowledgement that validity of biomarkers (e.g., their predictive value) needs to be defined.

Disease biomarkers

Herein, we define a dyscognitive state as a state that predicts cognitive impairment based on an underlying defect such as a mutation in a specific gene. It is useful when considering

"disease biomarkers" versus "symptomatic biomarkers," where the disease biomarker is a dyscognitive state and the symptomatic biomarker is the symptom (Table 4).

For example, a mutation in MeCP2 gene predicts a dyscognitive state in Rett syndrome. This prediction is useful even if it is employed in the presymptomatic stages. In this case the mutation is a biomarker predicting a dyscognitive state (Chahrour & Zoghbi, 2007; Samaco et al., 2008). In AD, a biomarker detecting amyloid deposits would be a diagnostic tool for determination of dyscognitive state. Examples include in vivo imaging with ^{11}C-PiB-PET amyloid (Driscoll et al., 2012). In patients with mild cognitive impairment (MCI), an increased ratio of $t_{tau}/A\beta_{1–42}$ (total tau protein/Aβ 1–42) in the cerebrospinal fluid (CSF) may be a biomarker that predicts a dyscognitive state and also predicts progression in AD (Shaw et al., 2011).

Symptomatic biomarkers

Symptomatic biomarkers reflect clinical manifestations of the comorbidity, without necessarily affecting the underlying disease For instance, status epilepticus predicts postictal cognitive impairment. Likewise, electrographic signatures of epileptic encephalopathies associated with cognitive dysfunction are symptomatic biomarkers. Electrical status epilepticus in sleep is a symptomatic biomarker of Landau-Kleffner syndrome (Galanopoulou et al., 2000; Tassinari et al., 2009).

Biomarkers for treatment implementation

Specific electrographic abnormalities in electrographic status epilepticus or epileptic encephalopathies of infancy may serve as biomarkers that can be used to define important variables. For example, these biomarkers help determine a therapeutic window, because their presence or absence defines efficacy of treatment. They also help guide treatment. Examples include the EEG-guided implementation of barbiturates in status epilepticus (Rossetti & Lowenstein, 2011), hormonal treatments in infantile spasms, or treatment for electrical status epilepticus in sleep (Galanopoulou et al., 2000; Pellock et al., 2010). Another example where biomarkers are used for treatment implementation is the information provided by diagnostic tests to localize the tumor responsible for secreting autoantibodies in autoimmune-mediated paraneoplastic syndromes manifesting in epilepsy (Rosenfeld & Dalmau, 2011).

Infrastructure

There are four areas that need to be addressed regarding infrastructure/resources for studies of the comorbidities of the epilepsies. They are (1) validated animal models, (2) training, (3) technology, and (4) funding. First, it is critical to utilize validated animal models for research on the comorbidities of the epilepsies. These models should have characteristics of the human condition they are modeling, and there should be an understanding of the seizure types and epilepsy syndrome that the animals simulate. It is critical that researchers improve the rigor of preclinical studies to maximize the likelihood of success in clinical trials (Landis et al., 2012). In addition, recommendations should be provided for validated protocols so that it is possible to compare results across animal models and laboratories.

Technological advances could greatly enhance research. It would be extremely helpful to develop automated methods to test animals that are less time-intensive, and to develop technology that facilitates long-term EEG monitoring. Developing technology to permit in vivo monitoring of biomarkers would be extremely useful.

There is a significant need for development of interdisciplinary research teams. Epilepsy research teams need to have access to investigators with expertise in testing rodent models and increase training for those who are directly involved in testing the animals.

This research would benefit from an increase in the availability of funds for studies on the comorbidities of the epilepsies, as well as increased interest for support of this type of research by more funding agencies.

Conclusion

Comorbidities in epilepsy can be debilitating, but they are often overlooked. Fortunately, a number of recent efforts are increasing the attention paid to comorbidities. Collaborations between diverse groups with specialized expertise—ranging from cognitive neuroscientists to clinical epileptologists—are making it possible to identify comorbidities earlier, define them more precisely, and develop new treatments. As these new efforts are underway, it will be essential to use terms, methods, and strategies clearly and consistently. Here we have suggested terminology, guidelines for methods, and outline approaches that will help the epilepsy community advance our understanding, diagnostics, and treatment of comorbidities.

Acknowledgments

The authors would like to thank the American Epilepsy Society (AES), International League Against Epilepsy (ILAE), Citizens United for Research in Epilepsy (CURE), Epilepsy Therapy Project, and Autism Speaks for their financial support of the AES/ILAE Joint workshop in London (September 2012). The authors would also like to thank Patricia Lear and Jennifer Pearson for their invaluable assistance in preparation of this manuscript.

Funding sources: R01NS38595 and grants from CURE, AES, the Epilepsy Foundation (EF) and the Colorado Center for Drug Discovery (ARB-K); NARSAD and EF (KGB); R37NS31146 and a grant from the Pediatric Epilepsy Foundation (ATB); R21NS078333 and R01NS020253 and grants from Autism Speaks, CURE, Heffer and Siegel Families Foundations (ASG); R01NS061991 and CURE (MRW); RO1NS039587, CURE, and Office of the Director grant R21NS072099 (MP); R01NS37562, R21MH090606, the Alzheimer's Association, and NYS Office of Mental Health (HES).

References

Adelow C, Andersson T, Ahlbom A, Tomson T. Hospitalization for psychiatric disorders before and after onset of unprovoked seizures/epilepsy. Neurology. 2012; 78:396–401. [PubMed: 22282649]

Aldenkamp AP, Alpherts WC, Dekker MJ, Overweg J. Neuropsychological aspects of learning disabilities in epilepsy. Epilepsia. 1990; 31(Suppl 4):S9–S20. [PubMed: 2279487]

American Psychiatric Association. Diagnostic and statistical manual of mental disorders DSM-IV-TR. 4. American Psychiatric Publishing; Arlington, VA: 2000.

Anisman H, McIntyre DC. Conceptual, spatial, and cue learning in the Morris water maze in fast or slow kindling rats: attention deficit comorbidity. J Neurosci. 2002; 22:7809–7817. [PubMed: 12196604]

Bailet LL, Turk WR. The impact of childhood epilepsy on neurocognitive and behavioral performance: a prospective longitudinal study. Epilepsia. 2000; 41:426–431. [PubMed: 10756408]

Beerpoot LJ, Lipska BK, Weinberger DR. Neurobiology of treatment-resistant schizophrenia: new insights and new models. Eur Neuropsychopharmacol. 1996; 6(Suppl 2):S27–S34. [PubMed: 8792118]

Berg AT, Langfitt JT, Testa FM, Levy SR, DiMario F, Westerveld M, Kulas J. Global cognitive function in children with epilepsy: a community-based study. Epilepsia. 2008; 49:608–614. [PubMed: 18070088]

Berg AT, Berkovic SF, Brodie MJ, Buchhalter J, Cross JH, van Emde BW, Engel J, French J, Glauser TA, Mathern GW, Moshé SL, Nordli D, Plouin P, Scheffer IE. Revised terminology and concepts

for organization of seizures and epilepsies: report of the ilae commission on classification and terminology, 2005–2009. Epilepsia. 2010; 51:676–685. [PubMed: 20196795]

Berg AT, Hesdorffer DC, Zelko FA. Special education participation in children with epilepsy: what does it reflect? Epilepsy Behav. 2011; 22:336–341. [PubMed: 21849261]

Biomarkers Definitions Working Group. Biomarkers and surrogate endpoints: preferred definitions and conceptual framework. Clin Pharmacol Ther. 2001; 69:89–95. [PubMed: 11240971]

Blumenfeld H, Klein JP, Schridde U, Vestal M, Rice T, Khera DS, Bashyal C, Giblin K, Paul-Laughinghouse C, Wang F, Phadke A, Mission J, Agarwal RK, Englot DJ, Motelow J, Nersesyan H, Waxman SG, Levin AR. Early treatment suppresses the development of spike-wave epilepsy in a rat model. Epilepsia. 2008; 49:400–409. [PubMed: 18070091]

Bolanos AR, Sarkisian M, Yang Y, Hori A, Helmers SL, Mikati M, Tandon P, Stafstrom CE, Holmes GL. Comparison of valproate and phenobarbital treatment after status epilepticus in rats. Neurology. 1998; 51:41–48. [PubMed: 9674776]

Bouilleret V, Hogan RE, Velakoulis D, Salzberg MR, Wang L, Egan GF, O'Brien TJ, Jones NC. Morphometric abnormalities and hyperanxiety in genetically epileptic rats: a model of psychiatric comorbidity? Neuroimage. 2009; 45:267–274. [PubMed: 19167503]

Cammisuli S, Murphy MP, Ikeda-Douglas CJ, Balkissoon V, Holsinger RM, Head E, Michael M, Racine RJ, Milgram NW. Effects of extended electrical kindling on exploratory behavior and spatial learning. Behav Brain Res. 1997; 89:179–190. [PubMed: 9475625]

Caroni P, Donato F, Muller D. Structural plasticity upon learning: regulation and functions. Nat Rev Neurosci. 2012; 13:478–490. [PubMed: 22714019]

Catterall WA, Kalume F, Oakley JC. Nav1.1 channels and epilepsy. J Physiol. 2010; 588:1849–1859. [PubMed: 20194124]

Chahrour M, Zoghbi HY. The story of rett syndrome: from clinic to neurobiology. Neuron. 2007; 56:422–437. [PubMed: 17988628]

Chang YC, Huang AM, Kuo YM, Wang ST, Chang YY, Huang CC. Febrile seizures impair memory and cAMP response-element binding protein activation. Ann Neurol. 2003; 54:706–718. [PubMed: 14681880]

Chauviere L, Rafrafi N, Thinus-Blanc C, Bartolomei F, Esclapez M, Bernard C. Early deficits in spatial memory and theta rhythm in experimental temporal lobe epilepsy. J Neurosci. 2009; 29:5402–5410. [PubMed: 19403808]

Cilio MR, Sogawa Y, Cha BH, Liu X, Huang LT, Holmes GL. Long-term effects of status epilepticus in the immature brain are specific for age and model. Epilepsia. 2003; 44:518–528. [PubMed: 12681000]

David DJ, Klemenhagen KC, Holick KA, Saxe MD, Mendez I, Santarelli L, Craig DA, Zhong H, Swanson CJ, Hegde LG, Ping XI, Dong D, Marzabadi MR, Gerald CP, Hen R. Efficacy of the mchr1 antagonist N-[3-(1-{[4-(3,4-difluorophenoxy)phenyl]methyl}(4-piperidyl))-4-methylphenyl]-2-m ethylpropanamide (SNAP 94847) in mouse models of anxiety and depression following acute and chronic administration is independent of hippocampal neurogenesis. J Pharmacol Exp Ther. 2007; 321:237–248. [PubMed: 17237257]

Davies S, Heyman I, Goodman R. A population survey of mental health problems in children with epilepsy. Dev Med Child Neurol. 2003; 45:292–295. [PubMed: 12729141]

de Rogalski Landrot I, Minokoshi M, Silveira DC, Cha BH, Holmes GL. Recurrent neonatal seizures: relationship of pathology to the electroencephalogram and cognition. Dev Brain Res. 2001; 129:27–38. [PubMed: 11454410]

Detour J, Schroeder H, Desor D, Nehlig A. A 5-month period of epilepsy impairs spatial memory, decreases anxiety, but spares object recognition in the lithium-pilocarpine model in adult rats. Epilepsia. 2005; 46:499–508. [PubMed: 15816943]

Driscoll I, Troncoso JC, Rudow G, Sojkova J, Pletnikova O, Zhou Y, Kraut MA, Ferrucci L, Mathis CA, Klunk WE, O'Brien RJ, Davatzikos C, Wong DF, Resnick SM. Correspondence between in vivo (11)c-pib-pet amyloid imaging and postmortem, region-matched assessment of plaques. Acta Neuropathol. 2012; 124:823–831. [PubMed: 22864813]

Dube CM, Zhou JL, Hamamura M, Zhao Q, Ring A, Abrahams J, McIntyre K, Nalcioglu O, Shatskih T, Baram TZ, Holmes GL. Cognitive dysfunction after experimental febrile seizures. Exp Neurol. 2009; 215:167–177. [PubMed: 19000675]

Ehninger D, Han S, Shilyansky C, Zhou Y, Li W, Kwiatkowski DJ, Ramesh V, Silva AJ. Reversal of learning deficits in a TSC2±mouse model of tuberous sclerosis. Nat Med. 2008; 14:843–848. [PubMed: 18568033]

Epps SA, Tabb KD, Lin SJ, Kahn AB, Javors MA, Boss-Williams KA, Weiss JM, Weinshenker D. Seizure susceptibility and epileptogenesis in a rat model of epilepsy and depression co-morbidity. Neuropsychopharmacology. 2012; 37:2756–2763. [PubMed: 22871911]

Fastenau PS, Jianzhao S, Dunn DW, Austin JK. Academic underachievement among children with epilepsy: proportion exceeding psychometric criteria for learning disability and associated risk factors. J Learn Disabil. 2008; 41:195–207. [PubMed: 18434287]

Faverjon S, Silveira DC, Fu DD, Cha BH, Akman C, Hu Y, Holmes GL. Beneficial effects of enriched environment following status epilepticus in immature rats. Neurology. 2002; 59:1356–1364. [PubMed: 12427884]

de Feo MR, Mecarelli O, Palladini G, Ricci GF. Long-term effects of early status epilepticus on the acquisition of conditioned avoidance behavior in rats. Epilepsia. 1986; 27:476–482. [PubMed: 3757934]

Galanopoulou AS, Bojko A, Lado F, Moshe SL. The spectrum of neuropsychiatric abnormalities associated with electrical status epilepticus in sleep. Brain Dev. 2000; 22:279–295. [PubMed: 10891635]

Galanopoulou AS, Buckmaster PS, Staley KJ, Moshe SL, Perucca E, Engel J Jr, Loscher W, Noebels JL, Pitkanen A, Stables J, White HS, O'Brien TJ, Simonato M. Identification of new epilepsy treatments: issues in preclinical methodology. Epilepsia. 2012; 53:571–582. [PubMed: 22292566]

Getova D, Bowery NG, Spassov V. Effects of gabab receptor antagonists on learning and memory retention in a rat model of absence epilepsy. Eur J Pharmacol. 1997; 320:9–13. [PubMed: 9049596]

Gilbert TH, McNamara RK, Corcoran ME. Kindling of hippocampal field CA1 impairs spatial learning and retention in the morris water maze. Behav Brain Res. 1996; 82:57–66. [PubMed: 9021070]

Gilby KL, Thorne V, Patey A, McIntyre DC. Ruling out postnatal origins to attention-deficit/hyperactivity disorder (ADHD)-like behaviors in a seizure-prone rat strain. Behav Neurosci. 2007; 121:370–379. [PubMed: 17469927]

Gilliam F, Hecimovic H, Sheline Y. Psychiatric comorbidity, health, and function in epilepsy. Epilepsy Behav. 2003; 4(Suppl 4):S26–S30. [PubMed: 14654425]

Gobbo OL, O'Mara SM. Impact of enriched-environment housing on brain-derived neurotrophic factor and on cognitive performance after a transient global ischemia. Behav Brain Res. 2004a; 152:231–241. [PubMed: 15196790]

Gobbo OL, O'Mara SM. Post-treatment, but not pre-treatment, with the selective cyclooxygenase-2 inhibitor celecoxib markedly enhances functional recovery from kainic acid-induced neurodegeneration. Neuroscience. 2004b; 125:317–327. [PubMed: 15062975]

Groticke I, Hoffmann K, Loscher W. Behavioral alterations in the pilocarpine model of temporal lobe epilepsy in mice. Exp Neurol. 2007; 207:329–349. [PubMed: 17714705]

Groticke I, Hoffmann K, Loscher W. Behavioral alterations in a mouse model of temporal lobe epilepsy induced by intrahippocampal injection of kainate. Exp Neurol. 2008; 213:71–83. [PubMed: 18585709]

Hamm RJ, O'Dell DM, Pike BR, Lyeth BG. Cognitive impairment following traumatic brain injury: the effect of pre- and post-injury administration of scopolamine and mk-801. Brain Res Cogn Brain Res. 1993; 1:223–226. [PubMed: 8003921]

Hannesson DK, Corcoran ME. The mnemonic effects of kindling. Neurosci Biobehav Rev. 2000; 24:725–751. [PubMed: 10974354]

Hannesson DK, Mohapel P, Corcoran ME. Dorsal hippocampal kindling selectively impairs spatial learning/short-term memory. Hippocampus. 2001a; 11:275–286. [PubMed: 11769309]

Hannesson DK, Howland J, Pollock M, Mohapel P, Wallace AE, Corcoran ME. Dorsal hippocampal kindling produces a selective and enduring disruption of hippocampally mediated behavior. J Neurosci. 2001b; 21:4443–4450. [PubMed: 11404431]

Harrigan T, Peredery O, Persinger M. Radial maze learning deficits and mediodorsal thalamic damage in context of multifocal seizure-induced brain lesions. Behav Neurosci. 1991; 105:482–486. [PubMed: 1863369]

Hartman AL, Zheng X, Bergbower E, Kennedy M, Hardwick JM. Seizure tests distinguish intermittent fasting from the ketogenic diet. Epilepsia. 2010; 51:1395–1402. [PubMed: 20477852]

Hesdorffer DC, Ishihara L, Mynepalli L, Webb DJ, Weil J, Hauser WA. Epilepsy, suicidality, and psychiatric disorders: a bidirectional association. Ann Neurol. 2012; 72:184–191. [PubMed: 22887468]

Holmes GL. Effects of early seizures on later behavior and epileptogenicity. Ment Retard Dev Disabil Res Rev. 2004; 10:101–105. [PubMed: 15362164]

Holmes GL, Thompson JL, Marchi TA, Gabriel PS, Hogan MA, Carl FG, Feldman DS. Effects of seizures on learning, memory, and behavior in the genetically epilepsy-prone rat. Ann Neurol. 1990; 27:24–32. [PubMed: 2301924]

Hori A, Tandon P, Holmes GL, Stafstrom CE. Ketogenic diet: effects on expression of kindled seizures and behavior in adult rats. Epilepsia. 1997; 38:750–758. [PubMed: 9579901]

Hort J, Brozek G, Mares P, Langmeier M, Komarek V. Cognitive functions after pilocarpine-induced status epilepticus: changes during silent period precede appearance of spontaneous recurrent seizures. Epilepsia. 1999; 40:1177–1183. [PubMed: 10487179]

Hort J, Brozek G, Komarek V, Langmeier M, Mares P. Interstrain differences in cognitive functions in rats in relation to status epilepticus. Behav Brain Res. 2000; 112:77–83. [PubMed: 10862938]

Huang LT, Yang SN, Liou CW, Hung PL, Lai MC, Wang CL, Wang TJ. Pentylenetetrazol-induced recurrent seizures in rat pups: time course on spatial learning and long-term effects. Epilepsia. 2002; 43:567–573. [PubMed: 12060014]

Inostroza M, Cid E, Brotons-Mas J, Gal B, Aivar P, Uzcategui YG, Sandi C, Menendez de la Prida L. Hippocampal-dependent spatial memory in the water maze is preserved in an experimental model of temporal lobe epilepsy in rats. PLoS ONE. 2011; 6:e22372. [PubMed: 21829459]

Institutes of Medicine Committee on Public Health Dimensions of the Epilepsies. Epilepsy across the spectrum: promoting health and understanding. The National Academies Press; Washington, DC: 2012.

Jensen FE, Holmes GL, Lombroso CT, Blume HK, Firkusny IR. Age-dependent changes in long-term seizure susceptibility and behavior after hypoxia in rats. Epilepsia. 1992; 33:971–980. [PubMed: 1464280]

Jessberger S, Nakashima K, Clemenson GD Jr, Mejia E, Mathews E, Ure K, Ogawa S, Sinton CM, Gage FH, Hsieh J. Epigenetic modulation of seizure-induced neurogenesis and cognitive decline. J Neurosci. 2007; 27:5967–5975. [PubMed: 17537967]

Jhuang H, Garrote E, Mutch J, Yu X, Khilnani V, Poggio T, Steele AD, Serre T. Automated home-cage behavioral phenotyping of mice. Nat Commun. 2010; 1:68. [PubMed: 20842193]

Jobe, PC.; Weber, RJ. Co-morbidity of neurological and affective disorders: the model of the genetically epilepsy-prone rat. In: Gilliam, F.; Kanner, AM.; Sheline, YI., editors. Depression and brain dysfunction. Taylor and Francis Medical Books; London: 2005. p. 121-157.

Jobe PC, Picchioni AL, Chin L. Role of brain 5-hydroxytryptamine in audiogenic seizure in the rat. Life Sci. 1973; 13:1–13. [PubMed: 4270954]

Jobe PC, Dailey JW, Wernicke JF. A noradrenergic and serotonergic hypothesis of the linkage between epilepsy and affective disorders. Crit Rev Neurobiol. 1999; 13:317–356. [PubMed: 11028680]

Johnson MR, Shorvon SD. Heredity in epilepsy: neurodevelopment, comorbidity, and the neurological trait. Epilepsy Behav. 2011; 22:421–427. [PubMed: 21890419]

Jones NC, Cardamone L, Williams JP, Salzberg MR, Myers D, O'Brien TJ. Experimental traumatic brain injury induces a pervasive hyperanxious phenotype in rats. J Neurotrauma. 2008a; 25:1367–1374. [PubMed: 19061380]

Jones NC, Salzberg MR, Kumar G, Couper A, Morris MJ, O'Brien TJ. Elevated anxiety and depressive-like behavior in a rat model of genetic generalized epilepsy suggesting common causation. Exp Neurol. 2008b; 209:254–260. [PubMed: 18022621]

Jones NC, Kumar G, O'Brien TJ, Morris MJ, Rees SM, Salzberg MR. Anxiolytic effects of rapid amygdala kindling, and the influence of early life experience in rats. Behav Brain Res. 2009; 203:81–87. [PubMed: 19397932]

Jones NC, Martin S, Megatia I, Hakami T, Salzberg MR, Pinault D, Morris MJ, O'Brien TJ, van den Buuse M. A genetic epilepsy rat model displays endophenotypes of psychosis. Neurobiol Dis. 2010; 39:116–125. [PubMed: 20153428]

Kalynchuk LE. Long-term amygdala kindling in rats as a model for the study of interictal emotionality in temporal lobe epilepsy. Neurosci Biobehav Rev. 2000; 24:691–704. [PubMed: 10974352]

Kanner AM. Depression and epilepsy: a bidirectional relation? Epilepsia. 2011; 52(Suppl 1):21–27. [PubMed: 21214536]

Kanner A, Barry J, Gilliam F, Hermann B, Meador K. Anxiety disorders, sub-syndromic depressive episodes and major depressive episodes: do they differ on their impact on the quality of life of patients with epilepsy? Epilepsia. 2010; 51:1152–1158. [PubMed: 20477847]

Kemppainen EJ, Nissinen J, Pitkanen A. Fear conditioning is impaired in systemic kainic acid and amygdala-stimulation models of epilepsy. Epilepsia. 2006; 47:820–829. [PubMed: 16686646]

Kilkenny C, Browne WJ, Cuthill IC, Emerson M, Altman DG. Improving bioscience research reporting: the ARRIVE guidelines for reporting animal research. PLoS Biol. 2010; 8:e1000412. [PubMed: 20613859]

Kim S, Zhang S, Choi KH, Reister R, Do C, Baykiz AF, Gershenfeld HK. An e3 ubiquitin ligase, really interesting new gene (ring) finger 41, is a candidate gene for anxiety-like behavior and beta-carboline-induced seizures. Biol Psychiatry. 2009; 65:425–431. [PubMed: 18986647]

Kleen JK, Wu EX, Holmes GL, Scott RC, Lenck-Santini PP. Enhanced oscillatory activity in the hippocampal-prefrontal network is related to short-term memory function after early-life seizures. J Neurosci. 2011; 31:15397–15406. [PubMed: 22031886]

Koh S, Chung H, Xia H, Mahadevia A, Song Y. Environmental enrichment reverses the impaired exploratory behavior and altered gene expression induced by early-life seizures. J Child Neurol. 2005; 20:796–802. [PubMed: 16417873]

Koh S, Magid R, Chung H, Stine CD, Wilson DN. Depressive behavior and selective down-regulation of serotonin receptor expression after early-life seizures: reversal by environmental enrichment. Epilepsy Behav. 2007; 10:26–31. [PubMed: 17196883]

Konkle AT, McCarthy MM. Developmental time course of estradiol, testosterone, and dihydrotestosterone levels in discrete regions of male and female rat brain. Endocrinology. 2011; 152:223–235. [PubMed: 21068160]

Krsek P, Mikulecka A, Druga R, Kubova H, Hlinak Z, Suchomelova L, Mares P. Long-term behavioral and morphological consequences of nonconvulsive status epilepticus in rats. Epilepsy Behav. 2004; 5:180–191. [PubMed: 15123019]

Landis SC, Amara SG, Asadullah K, Austin CP, Blumenstein R, Bradley EW, Crystal RG, Darnell RB, Ferrante RJ, Fillit H, Finkelstein R, Fisher M, Gendelman HE, Golub RM, Goudreau JL, Gross RA, Gubitz AK, Hesterlee SE, Howells DW, Huguenard J, Kelner K, Koroshetz W, Krainc D, Lazic SE, Levine MS, Macleod MR, McCall JM, Moxley RT 3rd, Narasimhan K, Noble LJ, Perrin S, Porter JD, Steward O, Unger E, Utz U, Silberberg SD. A call for transparent reporting to optimize the predictive value of preclinical research. Nature. 2012; 490:187–191. [PubMed: 23060188]

Lee H, Dvorak D, Kao HY, Duffy AM, Scharfman HE, Fenton AA. Early cognitive experience prevents adult deficits in a neurodevelopmental schizophrenia model. Neuron. 2012; 75:714–724. [PubMed: 22920261]

Letty S, Lerner-Natoli M, Rondouin G. Differential impairments of spatial memory and social behavior in two models of limbic epilepsy. Epilepsia. 1995; 36:973–982. [PubMed: 7555961]

Leung LS, Zhao D, Shen B. Long-lasting effects of partial hippocampal kindling on hippocampal physiology and function. Hippocampus. 1994; 4:696–704. [PubMed: 7704112]

Lillrank SM, Lipska BK, Weinberger DR. Neurodevelopmental animal models of schizophrenia. Clin Neurosci. 1995; 3:98–104. [PubMed: 7583625]

Lin D, Boyle MP, Dollar P, Lee H, Lein ES, Perona P, Anderson DJ. Functional identification of an aggression locus in the mouse hypothalamus. Nature. 2011; 470:221–226. [PubMed: 21307935]

Lipska BK, Swerdlow NR, Geyer MA, Jaskiw GE, Braff DL, Weinberger DR. Neonatal excitotoxic hippocampal damage in rats causes post-pubertal changes in prepulse inhibition of startle and its disruption by apomorphine. Psychopharmacology. 1995; 122:35–43. [PubMed: 8711062]

Liu Z, Gatt A, Werner SJ, Mikati MA, Holmes GL. Long-term behavioral deficits following pilocarpine seizures in immature rats. Epilepsy Res. 1994; 19:191–204. [PubMed: 7698095]

Lynch M, Sayin U, Bownds J, Janumpalli S, Sutula T. Long-term consequences of early postnatal seizures on hippocampal learning and plasticity. Eur J Neurosci. 2000; 12:2252–2264. [PubMed: 10947804]

Majak K, Pitkänen A. Do seizures cause irreversible cognitive damage? Evidence from animal studies. Epilepsy Behav. 2004; 5(Suppl 1):S35–S44. [PubMed: 14725845]

Manno I, Macchi F, Caleo M, Bozzi Y. Environmental enrichment reduces spontaneous seizures in the q54 transgenic mouse model of temporal lobe epilepsy. Epilepsia. 2011; 52:e113–e117. [PubMed: 21762452]

Mazarati A, Shin D, Auvin S, Caplan R, Sankar R. Kindling epileptogenesis in immature rats leads to persistent depressive behavior. Epilepsy Behav. 2007; 10:377–383. [PubMed: 17368107]

Mazarati AM, Pineda E, Shin D, Tio D, Taylor AN, Sankar R. Comorbidity between epilepsy and depression: role of hippocampal interleukin-1beta. Neurobiol Dis. 2010; 37:461–467. [PubMed: 19900553]

McCarthy MM. How it's made: organisational effects of hormones on the developing brain. J Neuroendocrinol. 2010; 22:736–742. [PubMed: 20646174]

McCaughran JA, Manetto C, Schechter N. Long-term deficits in passive avoidance responding following experimental febrile convulsions during infancy. Behav Brain Res. 1982; 5:73–79. [PubMed: 7082468]

McIntyre DC, McLeod WS, Anisman H. Working and reference memory in seizure-prone and seizure-resistant rats: impact of amygdala kindling. Behav Neurosci. 2004; 118:314–323. [PubMed: 15113257]

Mesquita AR, Tavares HB, Silva R, Sousa N. Febrile convulsions in developing rats induce a hyperanxious phenotype later in life. Epilepsy Behav. 2006; 9:401–406. [PubMed: 16950656]

Metz-Lutz MN, Kleitz C, de Saint MA, Massa R, Hirsch E, Marescaux C. Cognitive development in benign focal epilepsies of childhood. Dev Neurosci. 1999; 21:182–190. [PubMed: 10575241]

Mikati MA, Zeinieh MP, Kurdi RM, Harb SA, El Hokayem JA, Daderian RH, Shamseddine A, Obeid M, Bitar FF, El Sabban M. Long-term effects of acute and of chronic hypoxia on behavior and on hippocampal histology in the developing brain. Dev Brain Res. 2005; 157:98–102. [PubMed: 15939090]

Mikulecka A, Krsek P, Mares P. Nonconvulsive kainic acid-induced seizures elicit age-dependent impairment of memory for the elevated plus-maze. Epilepsy Behav. 2000; 1:418–426. [PubMed: 12737831]

Milman A, Rosenberg A, Weizman R, Pick CG. Mild traumatic brain injury induces persistent cognitive deficits and behavioral disturbances in mice. J Neurotrauma. 2005; 22:1003–1010. [PubMed: 16156715]

Minkeviciene R, Rheims S, Dobszay MB, Zilberter M, Hartikainen J, Fulop L, Penke B, Zilberter Y, Harkany T, Pitkanen A, Tanila H. Amyloid beta-induced neuronal hyperexcitability triggers progressive epilepsy. J Neurosci. 2009; 29:3453–3462. [PubMed: 19295151]

Mohajeri MH, Saini K, Li H, Crameri A, Lipp HP, Wolfer DP, Nitsch RM. Intact spatial memory in mice with seizure-induced partial loss of hippocampal pyramidal neurons. Neurobiol Dis. 2003; 12:174–181. [PubMed: 12742738]

Mula M. Epilepsy: bidirectional link between epilepsy and psychiatric disorders. Nat Rev Neurol. 2012; 8:252–253. [PubMed: 22508232]

Nissinen J, Halonen T, Koivisto E, Pitkanen A. A new model of chronic temporal lobe epilepsy induced by electrical stimulation of the amygdala in rat. Epilepsy Res. 2000; 38:177–205. [PubMed: 10642046]

Okaichi Y, Amano S, Ihara N, Hayase Y, Tazumi T, Okaichi H. Open-field behaviors and water-maze learning in the F substrain of Ihara epileptic rats. Epilepsia. 2006; 47:55–63. [PubMed: 16417532]

Oostrom K, Smeets-Schouten A, Kruitwagen C, Peters A, Jennekens-Schinkel A. Not only a matter of epilepsy: early problems of cognition and behavior in children with “epilepsy only”–a prospective, longitudinal, controlled study starting at diagnosis. Pediatrics. 2003; 112:1338–1344. [PubMed: 14654607]

Oostrom K, van Teeseling H, Smeets-Schouten A, Peters A, Jennekens-Schinkel A. Three to four years after diagnosis: cognition and behavior in children with ‘epilepsy only’. A prospective, controlled study. Brain. 2005; 128:1546–1555. [PubMed: 15817514]

Ott D, Siddarth P, Gurbani S, Koh S, Tournay A, Shields WD, Caplan R. Behavioral disorders in pediatric epilepsy: unmet psychiatric need. Epilepsia. 2003; 44:591–597. [PubMed: 12681010]

Palop JJ, Chin J, Roberson ED, Wang J, Thwin MT, Bien-Ly N, Yoo J, Ho KO, Yu GQ, Kreitzer A, Finkbeiner S, Noebels JL, Mucke L. Aberrant excitatory neuronal activity and compensatory remodeling of inhibitory hippocampal circuits in mouse models of alzheimer’s disease. Neuron. 2007; 55:697–711. [PubMed: 17785178]

Peele DB, Gilbert ME. Functional dissociation of acute and persistent cognitive deficits accompanying amygdala-kindled seizures. Behav Brain Res. 1992; 48:65–76. [PubMed: 1622555]

Pellock JM, Hrachovy R, Shinnar S, Baram TZ, Bettis D, Dlugos DJ, Gaillard WD, Gibson PA, Holmes GL, Nordl DR, O’Dell C, Shields WD, Trevathan E, Wheless JW. Infantile spasms: a U.S. consensus report. Epilepsia. 2010; 51:2175–2189. [PubMed: 20608959]

Pineda E, Shin D, Sankar R, Mazarati AM. Comorbidity between epilepsy and depression: experimental evidence for the involvement of serotonergic, glucocorticoid, and neuroinflammatory mechanisms. Epilepsia. 2010; 51(Suppl 3):110–114. [PubMed: 20618414]

Prendiville S, Gale K. Anticonvulsant effect of fluoxetine on focally evoked limbic motor seizures in rats. Epilepsia. 1993; 34:381–384. [PubMed: 8384110]

Rice AC, Floyd CL, Lyeth BG, Hamm RJ, DeLorenzo RJ. Status epilepticus causes long-term NMDA receptor-dependent behavioral changes and cognitive deficits. Epilepsia. 1998; 39:1148–1157. [PubMed: 9821978]

Rigor in Science Workgroup N. Improving the quality of NINDS-supported preclinical and clinical research through rigorous study design and transparent reporting. 2011. www.ninds.nih.gov/funding/transparency_in_reporting_guidance.pdf

Rosenfeld MR, Dalmau J. Anti-nmda-receptor encephalitis and other synaptic autoimmune disorders. Curr Treat Options Neurol. 2011; 13:324–332. [PubMed: 21298406]

Rossetti AO, Lowenstein DH. Management of refractory status epilepticus in adults: still more questions than answers. Lancet Neurol. 2011; 10:922–930. [PubMed: 21939901]

Runke D, McIntyre DC, St-Onge V, Gilby KL. Relation between startle reactivity and sucrose avidity in two rat strains bred for differential seizure susceptibility. Exp Neurol. 2011; 229:259–263. [PubMed: 21324316]

Russo E, Citraro R, Scicchitano F, De Fazio S, Perrota I, Di Paola ED, Constanti A, De Sarro G. Effects of early long-term treatment with antiepileptic drugs on development of seizures and depressive-like behavior in a rat genetic absence epilepsy model. Epilepsia. 2011; 52:1341–1350. [PubMed: 21635238]

Rutten A, van Albada M, Silveira DC, Cha BH, Liu X, Hu YN, Cilio MR, Holmes GL. Memory impairment following status epilepticus in immature rats: time-course and environmental effects. Eur J Neurosci. 2002; 16:501–513. [PubMed: 12193194]

Samaco RC, Fryer JD, Ren J, Fyffe S, Chao HT, Sun Y, Greer JJ, Zoghbi HY, Neul JL. A partial loss of function allele of methyl-cpg-binding protein 2 predicts a human neurodevelopmental syndrome. Hum Mol Genet. 2008; 17:1718–1727. [PubMed: 18321864]

Sarkisova KY, Kulikov MA. Behavioral characteristics of WAG/rij rats susceptible and non-susceptible to audiogenic seizures. Behav Brain Res. 2006; 166:9–18. [PubMed: 16183145]

Sarkisova K, van Luijtelaar G. The WAG/rij strain: a genetic animal model of absence epilepsy with comorbidity of depression. Prog Neuropsychopharmacol Biol Psychiatry. 2011; 35:854–876. [PubMed: 21093520]

Sarkisova KY, Midzianovskaia IS, Kulikov MA. Depressive-like behavioral alterations and c-fos expression in the dopaminergic brain regions in WAG/rij rats with genetic absence epilepsy. Behav Brain Res. 2003; 144:211–226. [PubMed: 12946611]

Sayin U, Sutula TP, Stafstrom CE. Seizures in the developing brain cause adverse long-term effects on spatial learning and anxiety. Epilepsia. 2004; 45:1539–1548. [PubMed: 15571512]

Scharfman HE. Untangling epilepsy and Alzheimer's disease. Epilepsy Curr. 2012a; 12:178–183. [PubMed: 23118602]

Scharfman HE. Alzheimer's disease and epilepsy: insight from animal models. Future Neurol. 2012b; 7:177–192. [PubMed: 22723738]

Shaw LM, Vanderstichele H, Knapik-Czajka M, Figurski M, Coart E, Blennow K, Soares H, Simon AJ, Lewczuk P, Dean RA, Siemers E, Potter W, Lee VM, Trojanowski JQ. Qualification of the analytical and clinical performance of csf biomarker analyses in adni. Acta Neuropathol. 2011; 121:597–609. [PubMed: 21311900]

Shimomura C, Ohta H. Behavioral abnormalities and seizure susceptibility in rat after neonatal anoxia. Brain Dev. 1988; 10:160–163. [PubMed: 3407852]

Sogawa Y, Monokoshi M, Silveira DC, Cha BH, Cilio MR, McCabe BK, Liu X, Hu Y, Holmes GL. Timing of cognitive deficits following neonatal seizures: relationship to histological changes in the hippocampus. Dev Brain Res. 2001; 131:73–83. [PubMed: 11718838]

Sroubek J, Hort J, Komarek V, Langmeier M, Brozek G. Acquisition and retrieval of conditioned taste aversion is impaired by brain damage caused by two hours of pilocarpine-induced status epilepticus. Physiol Res. 2001; 50:609–617. [PubMed: 11829323]

Stafstrom CE, Sasaki-Adams DM. NMDA-induced seizures in developing rats cause long-term learning impairment and increased seizure susceptibility. Epilepsy Res. 2003; 53:129–137. [PubMed: 12576174]

Stewart L, Bercovici E, Shukla R, Serbanescu I, Persad V, Mistry N, Cortez M, Snead OC 3rd. Daily rhythms of seizure activity and behavior in a model of atypical absence epilepsy. Epilepsy Behav. 2006; 9:564–572. [PubMed: 17030024]

Sutula T, Lauersdorf S, Lynch M, Jurgella C, Woodard A. Deficits in radial arm maze performance in kindled rats: evidence for long-lasting memory dysfunction induced by repeated brief seizures. J Neurosci. 1995; 15:8295–8301. [PubMed: 8613762]

Tabb K, Boss-Williams KA, Weiss JM, Weinshenker D. Rats bred for susceptibility to depression-like phenotypes have higher kainic acid-induced seizure mortality than their depression-resistant counterparts. Epilepsy Res. 2007; 74:140–146. [PubMed: 17400428]

Takechi K, Suemaru K, Kawasaki H, Araki H. Regulatory role of the dopamine and norepinephrine transporters in pentylenetetrazol-kindled mice: association with effect of antidepressants. Eur J Pharmacol. 2011; 673:33–39. [PubMed: 22044920]

Talos DM, Sun H, Zhou X, Fitzgerald EC, Jackson MC, Klein PM, Lan VJ, Joseph A, Jensen FE. The interaction between early life epilepsy and autistic-like behavioral consequences: a role for the mammalian target of rapamycin (mTOR) pathway. PLoS ONE. 2012; 7:e35885. [PubMed: 22567115]

Tassinari CA, Cantalupo G, Rios-Pohl L, Giustina ED, Rubboli G. Encephalopathy with status epilepticus during slow sleep: "The penelope syndrome". Epilepsia. 2009; 50(Suppl 7):4–8. [PubMed: 19682041]

Thio L, Rensing N, Maloney S, Wozniak D, Xiong C, Yamada K. A ketogenic diet does not impair rat behavior or long-term potentiation. Epilepsia. 2010; 51:1619–1623. [PubMed: 20132289]

van Eeghen AM, Black ME, Pulsifer MB, Kwiatkowski DJ, Thiele EA. Genotype and cognitive phenotype of patients with tuberous sclerosis complex. Eur J Hum Genet. 2012a; 20:510–515. [PubMed: 22189265]

van Eeghen AM, Pulsifer MB, Merker VL, Neumeyer AM, van Eeghen EE, Thibert RL, Cole AJ, Leigh FA, Plotkin SR, Thiele EA. Understanding relationships between autism, intelligence, and

epilepsy: a cross-disorder approach. Dev Med Child Neurol. 2012b; 55:146–153. [PubMed: 23205844]

Verret L, Mann EO, Hang GB, Barth AM, Cobos I, Ho K, Devidze N, Masliah E, Kreitzer AC, Mody I, Mucke L, Palop JJ. Inhibitory interneuron deficit links altered network activity and cognitive dysfunction in alzheimer model. Cell. 2012; 149:708–721. [PubMed: 22541439]

Wig GS, Barnes SJ, Pinel JP. Conditioning of a flavor aversion in rats by amygdala kindling. Behav Neurosci. 2002; 116:347–350. [PubMed: 11996319]

Wilhelm EA, Souza AC, Gai BM, Chagas PM, Roehrs JA, Nogueira CW. Hyperthermic seizures enhance responsiveness to pentylenetetrazole and induce cognitive dysfunction: protective effect of 3-alkynyl selenophene. Life Sci. 2012; 90:666–672. [PubMed: 22483695]

Xu K, Sun X, Eroku B, Tsipis C, Puchowicz M, LaManna JC. Diet-induced ketosis improves cognitive performance in aged rats. Adv Exp Med Biol. 2010; 662:71–75. [PubMed: 20204773]

Yan Q, Jobe P, Dailey JW. Further evidence of anticonvulsant role for 5-hydroxytryptamine in genetically epilepsy prone rats. Br J Pharmacol. 1995; 115:1314–1318. [PubMed: 7582562]

Yang Y, Liu Z, Cermak JM, Tandon P, Sarkisian MR, Stafstrom CE, Neill JC, Blusztajn JK, Holmes GL. Protective effects of prenatal choline supplementation on seizure-induced memory impairment. J Neurosci. 2000; 20:RC109. [PubMed: 11069978]

Yang L, Li F, Zhang H, Ge W, Mi C, Sun R, Liu C. Astrocyte activation and memory impairment in the repetitive febrile seizures model. Epilepsy Res. 2009; 86:209–220. [PubMed: 19643577]

Young D, Lawlor P, Leone P, Dragunow M, During M. Environmental enrichment inhibits spontaneous apoptosis, prevents seizures and is neuroprotective. Nat Med. 1999; 5:448–453. [PubMed: 10202938]

Zhao Q, Stafstrom C, Fu D, Hu Y, Holmes G. Detrimental effects of the ketogenic diet on cognitive function in rats. Pediatr Res. 2004; 55:498–506. [PubMed: 14711901]

Table 1

Examples of studies using common animal models of epilepsy to address comorbidities of epilepsy

Type of agent	Age when treated	Strain	Sex	Test	Delay	Results	References
I. Induction of SE							
Flurothyl	P0	Wistar	M	MWM open field	1 mo	Impaired less active (anxious)	Holmes, 2004
Flurothyl	P0–P12	Sprague	?	MWM	~1 mo	Impaired	de Rogalski Landrot et al., 2001
Flurothyl	P0–P9	Sprague	M	MWM	~1 mo	Impaired	Sogawa et al., 2001
Kainic acid	P1–24	?	M	RAM MWM Elevated plus	2–3 mo	Impaired Impaired Anxious	Sayin et al., 2004
Kainic acid	P1, P7, P14	Sprague	M	RAM	3–4 mo	Impaired	Lynch et al., 2000
Kainic acid or PTZ	P10, P25	Wistar	M	Active avoidance	~1 mo	Impaired	de Feo et al., 1986
Kainic acid	P18, P25	Wistar	M	Elevated plus	≤3 day	Impaired	Mikulecka et al., 2000
Kainic acid	P35	Sprague	M	MWM Handling test	~1 mo	Impaired Increased Emotionality	Bolanos et al., 1998
Kainic acid	Young adult	Wistar	M	Elevated plus	2 mo	No effect	Inostroza et al., 2011
Kainic acid	Young adult	Wistar	M	MWM	2 mo	No effect	Inostroza et al., 2011
Kainic acid	Young adult	Sprague	M	Elevated plus	2 mo	Less anxious	Inostroza et al., 2011
Kainic acid	Young adult	Sprague	M	MWM	2 mo	Impaired	Inostroza et al., 2011
Kainic acid	Adult	Fisher 344	F	Obj. Rec	~1 mo	Impaired	Jessberger et al., 2007
Kainic acid	Adult	Wistar	M	MWM Open field Obj. Rec	3 day–2 wk	Impaired Hyperactive Impaired	Gobbo & O'Mara, 2004a,b
Kainic acid into amygdala	Adult	Wistar	M	RAM	~1 mo	Impaired	Letty et al., 1995
Lithium-pilo	P20	Sprague	M	MWM	1 mo	Impaired	Faverjon et al., 2002
Lithium-pilo	Young adult	Sprague	M	Elevated plus	2 mo	Less anxious	Inostroza et al., 2011
Lithium-pilo	Young adult	Sprague	M	MWM	2 mo	Impaired	Inostroza et al., 2011
Lithium-pilo	Young adult	Wistar	M	Elevated plus	2 mo	Less anxious	Inostroza et al., 2011
Lithium-pilo	Young adult	Wistar	M	MWM	2 mo	Impaired	Inostroza et al., 2011
Lithium-pilo	Adult	Wistar	M	Social interaction Rotorod Elevated plus	2 mo	Impaired Impaired No effect	Krsek et al., 2004
Pilocarpine	Adult	Wistar	M	Object place Obj. rec	Varied	Impaired Impaired	Chauviere et al., 2009

Type of agent	Age when treated	Strain	Sex	Test	Delay	Results	References
Pilocarpine	P12, 16, & 20	Sprague	M	MWM	1–2 mo	Only P16 & P20 Impaired	Cilio et al., 2003
Pilocarpine	P20	Sprague	M	MWM	2 days to 1 mo	Impaired	Rutten et al., 2002a
Pilocarpine	P20, P45	Sprague	M	MWM	2–3 mo	Impaired	Liu et al., 1994
Pilocarpine	P34	Sprague	M	MWM	~1 wk	Impaired	Yang et al., 2000
Pilocarpine	Young Adult	C57bl/6, FVB	M/F	MWM	2 wk	Impaired	Mohajeri et al., 2003
Pilocarpine	Adult	Sprague	?	MWM	5 days, 6 wk	Impaired	Rice et al., 1998
Pilocarpine	Adult	Long-Evans	M	MWM	6 days	Impaired	Hort et al., 1999
Pilocarpine	Adult	Long-Evans Wistar	M	MWM	3–9 days	Impaired, worse In Wistar	Hort et al., 2000
Pilocarpine	Adult	Wistar	M	RAM	3 wk	Impaired	Harrigan et al., 1991
Pilocarpine	Adult	Long-Evans	M	Conditioned taste aversion	6–9 days	Impaired	Sroubek et al., 2001
PTZ	P10–14	Sprague	M	MWM Open field Rotarod	1 and 2 mo	Impaired No effect No effect	Huang et al., 2002
Amygdala stimulation	Adult	Sprague	M	MWM Fear conditioning	6 mo	Impaired Impaired	Nissinen et al., 2000
NMDA	P12–20	Sprague	M/F	MWM	2–3 mo	Impaired	Stafstrom & Sasaki-Adams, 2003
II. Kindling							
Amygdala	Adult	Long-Evans	M	Conditioned taste aversion	n/a	Increased consumption	Wig et al., 2002
Amygdala	Adult	Long-Evans	M	Conditioned taste aversion Passive avoidance	1–2 days	Impaired Impaired	Peele & Gilbert, 1992
Amygdala & perf. path	Adult	Long-Evans	M	MWM	~1 wk	No effect	Cammisuli et al., 1997
Olfactory bulb	Adult	Sprague	M	RAM	1 mo	Impaired	Sutula et al., 1995
Dorsal HC	Adult	Long-Evans	M	RAM	1, 3 wk	Impaired	Leung et al., 1994
Dorsal HC (CA1)	Adult	Long-Evans	M	MWM	24 *hr*[a]	Impaired Acquisition & Retention	Gilbert et al., 1996
Dorsal HC	Adult	Long-Evans	M	MWM	n/a	Impaired	Hannesson et al., 2001b
Dorsal HC	Adult	Long-Evans	M	MWM Obj. rec	7 days and 28 days	Impaired No effect	Hannesson et al., 2001a
Fast/slow kindled animals	Adult	Wistar	M	Variety of tasks	n/a	Spatial memory Working memory deficits Anxious Impulsive	Anisman & McIntyre, 2002; McIntyre et al., 2004; Runke et al., 2011
III. Brain injury							

Type of agent	Age when treated	Strain	Sex	Test	Delay	Results	References
FPI	P30 and Adult	Wistar	M	Sucrose preference Open field Elevated plus Forced swim	?	No effect Anxious Anxious No effect	Jones et al., 2008a
Mild TBI (weight drop)	Adult	Mice	M	Forced swim Swim T-maze Passive avoidance	7, 30, 60 and 90 days	Depressed 7 and 9 days Impaired only at 30d Impaired only at 30d	Milman et al., 2005
IV. Febrile							
Seizures							
SFS	P11	Sprague	M	MWM	3 mo	Impaired	Dube et al., 2009
SFS	P5, 10, 15 and 20	Sprague	M/F	Passive avoidance	~2 mo	Impaired except P5	McCaughran et al., 1982
SFS	P10	Wistar	M	MWM Elevated plus Forced swim Open field Tests of reflexes	3 mo	No effect Impaired (anxious) No effect Anxious No effect	Mesquita et al., 2006
SFS	P21	Wistar	M	Passive avoidance	1 mo	Impaired	Wilhelm et al., 2012
RFS	P10–12	Sprague	M/F	MWM Inh. avoidance	10 days	Impaired Impaired	Yang et al., 2009
RFS	P10–12	Sprague	M	MWM Inh. avoidance	~3 wk ~1 mo	Impaired Impaired	Chang, 2003
V. Neonatal							
Hypoxia							
Hypoxia	P10	Long-Evans	M	Open field Olfactory Habituation/Dishabituation Social choice test	Adult	No effect No effect Impaired	Talos et al.,2012
Hypoxia	P5, 10, or 60	Long-Evans	M	MWM Open field Handling	2–3 mo	Only P60 Impaired Only P60 Impaired Only P60 Impaired	Jensen et al., 1992
Anoxia	P4	Wistar	M/F	Open field Motor coordination Startle	P28, P56	Impaired P28 impaired P28 impaired	Shimomura & Ohta, 1988
Hypoxia	P10	Sprague	?	MWM Handling test	Adult	Impaired Increased aggression	Mikati et al., 2005
VI. Genetic							
GEPR	P30–35 and 45–75	GEPR	M	T-maze & MWM Open Field Intruder task Handling test	~3–5 mo	Impaired Less active Less aggressive More aggressive	Holmes et al., 1990

Type of agent	Age when treated	Strain	Sex	Test	Delay	Results	References
GAERS	Adult	GAERS, Wistar	M	Two-way active avoidance	n/a	GAERS better than nonepileptic control	Getova et al., 1997
GAERS	7 and 13 wk	GAERS	M/F	Sucrose preference Open field Elevated plus	n/a	Depressed Anxious Anxious	Jones et al., 2008b; Bouilleret et al., 2009
GAERS	6 and 14 wk	GAERS	M/F	Prepulse inhibition Startle Amphetamine-Induced locomotor test	n/a	No effect Increased Hyperactive	Jones et al., 2009
WAG/Rij	Adult	WAG/Rij	M	Open Field Elevated plus Sucrose preference Forced swim	3–4 wk After Audio. seizure	Anxious Anxious Depressed Depressed	Sarkisova et al., 2003; Sarkisova & Kulikov, 2006; Sarkisova & van Luijtelaar, 2011
Ihara, Wistar	6–12 wk and 7–9 mo	Ihara, Wistar	?	MWM Open field Cue task	n/a	Impaired Impaired Impaired	Okaichi et al., 2006

[a]Twenty-four hours after the last kindled seizure.

Studies are listed that conducted behavioral assays in animal models of epilepsy. Adapted from Majak & Pitkanen, 2004. The table is organized into three parts: Models where status epilepticus is used to induce epilepsy, models that use kindling, and other models. In each category the references that use young animals are listed first.

P, postnatal day; ?, information unavailable; mo, month; n/a, not applicable; MWM, Morris water maze; RAM, Radial arm maze; Obj. rec., Novel Object Recognition; Object place, Novel Object Placement; Inh. avoidance, inhibitory avoidance; PTZ, pentylenetetrazol; M, males; F, females; M/F, males and females; Sprague, Sprague-Dawley; FPI, fluid percussion injury; TBI, traumatic brain injury; SFS, single febrile seizure; RFS, repetitive febrile seizures.

Table 2

DSM-IV–adapted diagnostic criteria and evaluation of major depressive disorder and their use in rodents

I. DSM-IV criteria for major depressive disorder

- **A.** The following symptoms (A1 or A2 criteria are necessary, others are helpful but not required), and the demonstration of these symptoms for a sufficiently long period that they can be distinguished from previous functioning:
  - **1.** Depressed mood most of the day, nearly every day
  - **2.** Diminished interest or pleasure in all or almost all activities most of the day, nearly every day
  - **3.** Significant weight loss without dieting, or weight gain, or change in appetite, nearly every day
  - **4.** Insomnia or hypersomnia, nearly every day
  - **5.** Psychomotor agitation or retardation, nearly every day
  - **6.** Fatigue or loss of energy, nearly every day
  - **7.** Feelings of worthlessness, or excessive/inappropriate guilt, nearly every day (currently not testable in rodents)
  - **8.** Diminished ability to focus or concentrate, or indecisiveness, nearly every day
  - **9.** Recurrent thoughts of death or suicidal ideation without a specific plan or suicide attempt or specific plan to commit suicide
- **B.** Symptoms do not meet criteria for mixed episode
- **C.** Significant distress or impairment in social, occupational, or other important areas of functioning
- **D.** Symptoms are not due to drug effects or medical condition
- **E.** The symptoms are accounted for by bereavement

II. Proposed batteries of tests for DSM-IV–adapted criteria for depressive disorder in rodents

Test	Time to test	A1	A2	A3	A4	A5	A6	A8	A9	C
Homecage monitoring	Multiple sleep-wake cycles	X	X		X	X				X
Saccharin preference		X	X	X						
Porsolt swim		X								
Open field activity		X								
Social interaction										X
Body weight, food intake	Daily or regular intervals			X						
Novelty suppressed feeding				X						
Sleep monitoring	Multiple continuous sleep-wake cycles				X					
Tail suspension (mice only)		X				X				
5-Choice serial reaction								X		
Attentional set shifting								X		

Test	Time to test	A1	A2	A3	A4	A5	A6	A8	A9	C
Prepulse inhibition								X		
Spontaneous alternation								X		
Alternation										
Forced run							?			
Self-injury									X	

An example of a disorder, major depressive disorder, is used to illustrate the adaptation of DSM-IV criteria to a rodent, and behavioral tests appropriate for those criteria. I. A list of criteria based on DSM-IV that is consistent with major depressive disorder. II. A list of tests that can be used in rodents to establish the criteria in I. Note that several criteria in I are incompatible with research in rodents (e.g., A7, D, E).

Table 3

Common behavioral tests to evaluate cognitive and behavioral comorbidities in animal models of epilepsy

Comorbidity	Tests
Depression	Novelty suppressed feeding Porsolt forced swim task Saccharin preference test Social interaction Tail suspension task (mice only)
Anxiety	Defensive burying Elevated plus or Zero mazes Open field Social interaction
Cognitive dysfunction	Cued or contextual fear conditioning Contextual discrimination Delayed nonmatching to sample Morris water maze Novel object recognition, novel object placement, novel context Place preference task Radial arm maze Spontaneous alteration task
Sleep disorders	Circadian rhythm measurements (body temperature, corticosterone levels) Food and water intake Home cage monitoring Sleep EEG
Aggression	Isolation induced intruder-resident task Social interaction
Attention	Attentional set shifting task Five choice serial reaction task Prepulse inhibition
Sexual dysfunction	Estrous cyclicity Lordosis behavior Sexual receptivity task

Tests are listed that are useful in identifying a comorbidity (e.g., depression). Tests are listed in alphabetical order.

Table 4

Classification and types of biomarkers of comorbidities

Disease biomarkers	Symptomatic biomarkers	Biomarkers for treatment implementation
Predicting dyscognitive state	Predicting cognitive dysfunction	Biomarkers of therapeutic window
Diagnostic of dyscognitive state (by pathological or clinical criteria)	Diagnostic of cognitive dysfunction	Biomarkers of treatment efficacy Target engagement Efficacy Monitoring (drug levels, biologic effects)
Monitoring dyscognitive state (onset, progression, cure, recurrence)	Monitoring cognitive dysfunction (onset, progression, cure, recurrence)	Biomarkers that localize the underlying pathology Biomarkers of treatment toxicity

The table provides examples of potential biomarkers that are useful in predicting, diagnosing, or monitoring comorbidities. The biomarkers can reflect symptoms of the comorbidity or indicate therapeutic effects.