The Use of Animal Models for the Early Identification and Characterization of Anticonvulsant Efficacy

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The NINDS Anticonvulsant Screening Project

✓ NIH-sponsored AED Drug Discovery Program that encourages and facilitates the discovery and development of new AEDs

✓ Provides sponsors with early preclinical efficacy and toxicity data using a battery of animal models

✓ Partnership between SPONSOR, NIH, and THE UNIVERSITY OF UTAH.

✓ For more information, interested parties are encouraged to contact Mr. James P. Stables (stablesj@ninds.nih.gov) and consult the NINDS web-site:

http://ninds.nih.gov/funding/research/asp/index.htm
Mission Statement

The Anticonvulsant Screening Program (ASP) is a government sponsored effort to **encourage and to facilitate** the discovery of new therapeutic agents for the treatment of seizure disorders. The ASP has as its major goal the **establishment of worldwide collaborative relationships** among the government, academia, and industry to search for a cure of the epilepsies and to provide the necessary incentives for discovery, characterization, and development of novel antiseizure/anticonvulsant agents. These efforts are undertaken through **multilevel testing** directed toward the development of safer and more effective therapies for treating the various seizure disorders. The **biological information** generated and the **consultations provided** aid our participants in developing unique insights concerning the pharmacokinetic and pharmacodynamic properties of the compounds tested. ASP thus provides a strong incentive to researchers working in this and other fields. The success of these efforts directly translates into new **treatments** for patients afflicted with seizure disorders.
Current Era of AED Development

- Aimed at identifying drugs for the symptomatic treatment of epilepsy
- Employs well characterized animal seizure and epilepsy models
The “ideal” animal model should reflect human epilepsy

- Seizures should:
  - evolve spontaneously;
  - display a phenotype consistent with human epilepsy;
  - be therapy resistant; and
  - be amenable to high volume screening.

- Since the majority of human epilepsies are multi-factorial, a single animal model is unlikely.
<table>
<thead>
<tr>
<th>Animal Model</th>
<th>Seizure phenotype</th>
<th>Human correlate</th>
<th>Predictive validity</th>
<th>Pharmacology</th>
</tr>
</thead>
<tbody>
<tr>
<td>Maximal electroshock</td>
<td>Tonic-extension seizure</td>
<td>Generalized tonic-clonic seizures</td>
<td>Yes</td>
<td>Na+ channel blockers, K+ channel activators, NMDA and AMPA receptor antagonists, α2δ ligands</td>
</tr>
<tr>
<td>sc Metrazol</td>
<td>Minimal clonic seizure</td>
<td>Generalized myoclonic seizure</td>
<td>Yes (for the most part; e.g. Keppra)</td>
<td>T-type Ca^{2+} channel blockers, Benzodiazepines, Barbiturates, GABA transport blockers, GABA transaminase inhibitors, α2δ ligands</td>
</tr>
<tr>
<td>GAERS, Lethargic mouse, and Wistar rat</td>
<td>Spike-wave discharges</td>
<td>Generalized absence</td>
<td>Yes</td>
<td>T-type Ca^{2+} channel blockers, GABA_{A} receptor antagonists, SV2a ligands</td>
</tr>
<tr>
<td>Kindled rat</td>
<td>Limbic seizures 2° generalized</td>
<td>Partial seizures</td>
<td>Yes</td>
<td>Na+ channel blockers, K+ channel activators, AMPA receptor antagonists, GABA receptor modulators, SV2a ligands, α2δ ligands</td>
</tr>
</tbody>
</table>
### Clinical Utility and Efficacy in Animal Seizure and Epilepsy

<table>
<thead>
<tr>
<th>Animal model</th>
<th>GTC</th>
<th>Clinical Seizure Type</th>
<th>Myoclonic</th>
<th>Absence</th>
<th>Partial</th>
</tr>
</thead>
<tbody>
<tr>
<td>MES (tonic extension)</td>
<td>CBZ, PHT, VPA, PB</td>
<td>ESM, VPA, PB*, BZD</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>[FBM, GBP, LTG, TPM, ZNS]</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>sc PTZ (clonic seizure)</td>
<td>ESM, VPA, PB, BZD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>[FBM, GBP, TGB*, VGB*]</td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Spike-wave seizures</td>
<td>ESM, VPA, BZD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>[LTG, TPM, LVT]</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Kindling</td>
<td>CBZ, PHT, VPA, PB, BZD</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>[FBM, GBP, LTG, TPM, TGB, ZNS, LVT, VGB]</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

* PB, TGB, and VGB block clonic seizures induced by sc PTZ but are inactive against generalized absence seizures and may exacerbate spike wave seizures.
<table>
<thead>
<tr>
<th>Year</th>
<th>AED Name</th>
</tr>
</thead>
<tbody>
<tr>
<td>1993</td>
<td>Felbamate</td>
</tr>
<tr>
<td>1993</td>
<td>Gabapentin</td>
</tr>
<tr>
<td>1994</td>
<td>Lamotrigine</td>
</tr>
<tr>
<td>1996</td>
<td>Fosphenytoin</td>
</tr>
<tr>
<td>1996</td>
<td>Topiramate</td>
</tr>
<tr>
<td>1997</td>
<td>Tiagabine</td>
</tr>
<tr>
<td>1999</td>
<td>Levetiracetam</td>
</tr>
<tr>
<td>2000</td>
<td>Zonisamide</td>
</tr>
<tr>
<td>2000</td>
<td>Oxcarbazepine</td>
</tr>
<tr>
<td>2005</td>
<td>Pregabalin</td>
</tr>
</tbody>
</table>
More AEDs in the Pipeline

- Brivaracetam (ucb 34714) and Seletracetam (ucb 44212)
- Carisbamate (RWJ 333369 / YKP 509)
- Eslicarbazepine acetate (S-licarbazepine acetate, BIA 2-093)
- Fluorofelbamate
- Ganaxolone (CCD 1042)
- Huperzine A
- Isovaleramide (NPS 1776)
- JZP-4
- Lacosamide (harkoseride, SPM 927)
- Losigamone
- PID
- Retigabine (D23129)
- Rufinamide
- Safinamide (NW 1015, FCE 26743 PNU 151774)
- Stiripentol
- Talampanel (GYKI 53773; LY 300164)
- Valrocemide (TV-1901)
There remains a need for additional models

- Catastrophic Childhood Epilepsies
  - Neonatal seizures
  - Infantile spasms
  - Severe myoclonic epilepsies
  - Lennox-Gastaut Syndrome
- Post Traumatic Head Injury
- Stroke-Induced Epilepsy

Existing animal models do not predict efficacy for these conditions!
These 2 prospective studies of >1000 patients (1977-1992) were the basis of the statement that 30% of patients with partial-onset epilepsy are poorly controlled. This has not changed in 30 years!

One Drug (1st + 2nd) N=100

Typical Management N=70
Some Sz, Adverse Effects

Unsatisfactory -Change AED N=30
Excessive Sz, Adverse Effects

Two Drug Treatment N=30

Typical Management N=15
85% have some Sz, adverse effects

Unsatisfactory -Change AED N=15
Excessive Sz, Adverse Effects

Refractory to all 10%

Surgery 5%

Experimental AED (few)

© Cramer 1992
Current Models Do Not Appear To Be Addressing Pharmacoresistance

Newly treated epilepsy
n=470

1st AED
- Uncontrolled seizures 53%
- Seizure-free 47%

2nd AED
- Uncontrolled seizures 40%
- Seizure-free 13%

3rd AED
- Uncontrolled seizures 39%
- Seizure-free 1%

Polytherapy
- Uncontrolled seizures 36%
- Seizure-free 3%

What can (should) we be doing to identify more effective AEDs for the refractory patient population?

- Better define the mechanisms of pharmaco-resistant epilepsy
- Develop better models of refractory epilepsy
- Employ new models into drug discovery process
Refractory Epilepsy: A Multifactorial Process?

- Genetic polymorphisms
- Disease-related factors including:
  - etiology
  - progression with continued AED treatment
  - modification of receptors & or ion channels
  - seizure-induced synaptic reorganization
  - modified drug uptake into the brain
- Drug-related factors including:
  - pharmacodynamic tolerance
  - ineffective / inappropriate mechanism of existing drugs

Studies on tissue from drug-resistant patients and development of appropriate experimental models are clearly needed

Loscher and Schmidt, Epil. Res. 50: 3, 2002
Therapy Resistant Models

- Phenytoin-resistant kindled rat
- Lamotrigine-resistant kindled rat
- 6 Hz “Psychomotor” Seizures
- Post-status spontaneous seizure models
- In utero methylazoxymethanol (MAM) acetate-induced heterotopia
- In vitro brain slices from kainate-treated rats

Properties

- All six models display unique attributes that may model pharmacoresistant epilepsy
- Therapy resistant seizure models are more likely to identify novel AEDs

Yet to be “truly” validated by human experience
Laboratory Research
Lamotrigine Treatment During Amygdala-Kindled Seizure Development Fails to Inhibit Seizures and Diminishes Subsequent Anticonvulsant Efficacy

Terri Postma, *Eckart Krupp, Xiu-Li Li, Robert M. Post, and Susan R. B. Weiss

Biological Psychiatry Branch, National Institute of Mental Health, Bethesda, Maryland, U.S.A.
Pharmacology of the LTG-Resistant Kindled Rat

<table>
<thead>
<tr>
<th>Resistant</th>
<th>Sensitive</th>
</tr>
</thead>
<tbody>
<tr>
<td>✓ Lamotrigine</td>
<td>✓ Valproate</td>
</tr>
<tr>
<td>✓ Phenytoin</td>
<td>✓ Clonazepam</td>
</tr>
<tr>
<td>✓ Carbamazepine</td>
<td>✓ Levetiracetam</td>
</tr>
<tr>
<td>✓ Topiramate</td>
<td>✓ Carisbamate</td>
</tr>
<tr>
<td></td>
<td>✓ Retigabine</td>
</tr>
</tbody>
</table>

The LTG-kindled rat displays a pharmacological profile consistent with pharmaco-resistant epilepsy and is amenable to medium through-put screening.
6 Hz Corneal Stimulation Model

- Originally promoted as a model of “psychomotor” seizures (Brown et al., JPET, 107:273-283, 1953)
- **Stimulus**
  - 32 mA for 3 sec at 6 Hz
- **Seizure**
  - Stun followed by forelimb clonus, twitching of vibrissae, and Straub tail
- **Stimulus is thought to activate limbic brain structures**
## Pharmacology of 6 Hz Model

<table>
<thead>
<tr>
<th>AED</th>
<th>ED50 (mg/kg, i.p.) and 95% C.I. (^b)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>22 mA</td>
</tr>
<tr>
<td>Phenytoin</td>
<td>9.4</td>
</tr>
<tr>
<td>(4.7 - 14.9)</td>
<td></td>
</tr>
<tr>
<td>Lamotrigine</td>
<td>4.4</td>
</tr>
<tr>
<td>(2.2 - 6.6)</td>
<td></td>
</tr>
<tr>
<td>Ethosuximide</td>
<td>86.9</td>
</tr>
<tr>
<td>(37.8 - 156)</td>
<td>(114 - 223)</td>
</tr>
<tr>
<td>Levetiracetam</td>
<td>4.6</td>
</tr>
<tr>
<td>(1.1 - 8.7)</td>
<td>(9.9 - 36.0)</td>
</tr>
<tr>
<td>Valproic acid</td>
<td>41.5</td>
</tr>
<tr>
<td>(16.1 - 68.8)</td>
<td>(94.5 - 152)</td>
</tr>
</tbody>
</table>

Models of Pharmaco-resistant Epilepsy

- Available models display resistance to two or more of first-line AEDs
- Provide a useful means to “differentiate” investigational AEDs
What about AED testing in chronic seizure models

✓ Advantages
  – Animals display recurrent, spontaneous seizures
  – Histopathology similar to mesial temporal sclerosis
  – Animals have experienced epileptogenesis
Conclusions from Rat “Clinical Trials”

✓ Can distinguish efficacy between anticonvulsants
✓ May prove to be predictive for AEDs and the treatment of refractory epilepsy
✓ May help influence study design in clinic; e.g., seizure type, favorable AED combinations.

Not for the timid, but worth the effort!
Challenges for Future Drug Development

- Identify New Molecular Targets for Drugs

- Develop, Validate, and Implement New Model Systems into Drug Development
  - Intractable Epilepsy
  - Epileptogenesis
  - Mutant mouse models
  - Alternative approaches; i.e., HTS using Zebra fish and/or Drosophila systems

- Coordinate Clinical and Basic Research