Epilepsy & CBD

The Chain Ganglion:
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Meet Bubba!

Bubba is a 7 year old German Shepherd mix. He loves long walks, dinner time, and also...
...he is epileptic!
• Idiopathic epilepsy
  ○ ~60% of epilepsy is idiopathic
• On a regimen of KBr and Phenobarbital
• When he was first adopted he had seizures approximately every 2 weeks, which was much more frequent than we had been told to expect.
After a few months of the frequent seizures, we decided to start him on CBD and he improved almost immediately.

- His seizures became much less frequent (every 14-16 weeks)
- Shorter in duration with faster recovery
- Able to significantly lower the dose of other medications
  - Huge benefit because traditional epilepsy medications are hard on the liver
Ancient Outlook

- Historical perspective on seizures varies by culture
  - The Hammurabi Code (1780 BC) limited the rights of epileptics (marriage, oaths, as slaves); the hand of sin
  - Christian perspective of possession; infection through breath
- Stigma led to isolation furthering stereotypes
- Stigma present today in US laws
  - People with epilepsy were forbidden to marry in 17 states, until 1956
  - Last state repealed in 1980
What is epilepsy?

- Epilepsy is defined as a clinical syndrome with recurrent and unpredictable seizures. These seizures can take place without identifiable cause. A cause can only be found in about 1 in 100 people.
- Seen mostly in people who are over the age and 60 and young children.
- Seizures are moments of vigorous shaking and there is no guarantee how long one may last, this can sometimes lead to broken bones and other physical injuries.
Diagnosis of Epilepsy

- **Electroencephalogram (EEG)**
  - Used to record electrical activity on skull with 21 electrodes
  - Under normal conditions EEG activity in the brain will be low, but depending on the severity of the seizure the activity gets much bigger and more frequent

- **Imaging studies**
  - MRI and CT may be used to find location of a scar or damaged brain tissue.
  - PET scans may be used to examine blood flow
Causes of Epilepsy

The majority of epilepsy is *de novo* and idiopathic. Acquired epilepsy often has an identifiable cause.

- Genetics
- Structural abnormality
- Trauma
- Stroke
- Tumors
- Infectious disease
- Unknown causes
Glutamate and GABA

- GABA plays an inhibitory role
  - Allows $\text{Cl}^-$ into the cell and $\text{K}^+$ out of the cell causing hyperpolarization
  - $\text{GABA}_a$ ligand gated ion channel and $\text{GABA}_b$ a GPCR
- Glu plays an excitatory role
  - Increase membrane permeability for Ca, Na, K
  - AMPA receptors, NMDA receptors, and metabotropic glutamate receptors
- Decrease in GABA and increase in Glu change inhibitory/excitatory balance
Types of Seizures

- **Focal seizure**: abnormal electrical activity in only one area of the brain. Can spread and become a generalized seizure.
  - Simple focal
  - Complex focal

- **Generalized seizures**: discharge of neurons in both hemispheres. Most common type of epilepsy, starting in one area and spreading across the entire brain.
  - Tonic-clonic (grand mal)
  - Absence (petit mal)
  - Myoclonic
  - Atonic

- **Status epilepticus**: frequent and long seizures without regaining consciousness. Seizures >5 minutes, and results in 42,000 deaths per year in the US.
Epileptic Triggers

- Fever
- Substance withdrawal (alcohol/benzodiazepines)
- Flashing lights or sounds
- Low blood sugar
- Stress
- Lack of sleep
- Hormones

While triggers may induce seizure, they do NOT cause epilepsy
**Genetic Markers of Epilepsy**

MANY genetic markers of epilepsy exist, often indicating dysfunction of channels (channelopathies). General themes include:

- **Overactive Na\(^{+}\) or Ca\(^{2+}\) channels**
- **Inactive or underactive K\(^{+}\) channels**

A number of these genes are **implicated in other disorders**, and are related to other genes involving channel subunits.

Different mutations can cause different types of epilepsy.
SCN1A

Involved in the formation of Nav1.1 sodium channel (alpha subunit)

Implicated in febrile seizures, Dravet syndrome, and generalized tonic-clonic seizures

The type of mutation can determine the severity, ranging from febrile seizures to Dravet syndrome (Severe Myoclonic Epilepsy of Infancy)
SCN1A is very highly connected to many other genes which also code different protein subunits of sodium channels. Clearly, these linkages are well established in a variety of methods.

(CALM2 is Calmodulin 2, which mediates a large number of enzymes, ion channels, aquaporins and other proteins by Ca^{2+})
It’s related with high confidence to a number of genes apart from sodium channels, including other markers of epilepsy like **KCNN2** and **nitric oxide synthases** (which play a controversial role, seeming that overexpression of NOS is common in patients with epileptic history).
Epilepsy Comorbidity

- Migraine
- Bipolar Depression
- Anxiety
- Autism
- Alzheimer’s Disease
- (and more!)
Former and Current Treatments

● Former Treatments
  ○ Herbs
  ○ Surgery
    ■ Very invasive
    ■ Split brain surgery, temporal lobe surgery (patient HM), hemispherectomy
    ■ Large and variable side effects

● Current Treatments
  ○ Anticonvulsants
    ■ Work to increase GABA action, as Na+ channel blockers, and/or Ca2+ channel blockers
    ■ Going off medication can make seizures worse
  ○ Keto Diet
  ○ Biofeedback
  ○ Counterstimulation
  ○ Cannabidiol
Intro to CBD

- CBD is an abbreviation for cannabinoid
- CBD acts as an anti-inflammatory, anticonvulsant, and antioxidant agent
- CBD is being explored in its use to aid treatment of neuroinflammation, epilepsy, oxidative injury, vomiting, and more.

Epilepsy occurs when a flood of excitation creates incessant and abnormal firing of neurons. CBDs modulates the amount of neurotransmitter released, this keeps the brain stable by keeping neurotransmitter levels balanced.
Uses for CBD

- Relieves pain and inflammation
- Has antipsychotic effects
- Reduces anxiety
- May help fight cancer
- Relieves nausea, a useful antiemetic
- May treat seizures or other neurological disorders
- Lowers incidence of diabetes
Benefits of CBD for Epilepsy

CBD may affect epilepsy through a variety of mechanisms. Some of the pathways may be more direct effects of CBD on receptors and signaling, while others may be indirect mechanisms. Potential mechanisms include:

- **Direct inhibition of CBD on the hippocampus**
  - Via 5HT1A serotonergic autoreceptors?

- **Indirect activation of the endocannabinoid system**
  - Attenuates synaptic transmission
  - Induction of long term depression

- **Modulation of epileptic triggers**
  - Anxiolytic properties
  - Improved sleep
  - Hormonal repression

- **Alleviation of comorbid symptoms**
  - Anxiety, depression, migraine, bipolar, Alzheimer’s
Where CBD acts

- CBDs has a **high affinity at 5HT_{1A} receptors**
  - 5HT_{1A} receptors are GPCRs
- CBD has **low affinity at cannabinoid receptors**.
  - CB1 receptors are located mostly throughout the brain
  - CB2 receptors are located in the spleen and in other immune cells
Where CBD Acts cont.

Cannabidiol also **exerts an inhibitory effect on a variety of cytochrome p450 enzymes.** This is an important consideration in thinking about the effect of CBD in tandem with other medications as it may change drug metabolism.

- Does CBD actually control epilepsy extremely well?
- Does CBD just inhibit the breakdown of other medications, making lower doses more effective?
Indirect Activation of the Endocannabinoid System

CBD is a weak antagonist or negative allosteric modulator at CB1 and a weak agonist at CB2, however it also inhibits the reuptake and breakdown of anandamide, an endogenous cannabinoid which does activate CB receptors. CB1 receptors are prevalent in the CNS, and the relevant effects of their activation may include:

- Reducing activity at voltage gated calcium channels
- Attenuation of neurotransmitter release
- Induction of synaptic depression via TRPV1
- Modulation of hyper-corticosteroid secretion (HPA axis)
- Inhibition of excessive arousal in anxiety pathways
- Analgesia
- Increase in amount and quality of sleep
CB1 Distribution

- Expressed many places (most of the black represents mean amounts, not ‘no data’)
- Fairly similar expression to 5HT1A, but less in brainstem structures.
Epidiolex (cannabidiol)

- Patent No: US 9,474,726 B2
- Likely to be the first ever marijuana-derived medication ever approved by the FDA
- Federal health advisors panel unanimously in favor of the treatment at the meeting on April 19th, 2018
- FDA regulators due to make their decision in late June of this year
- GW Pharmaceuticals
  - Conducted three studies
  - Positive results from all three
Epidiolex

- Not meant to replace other CBD on the market, but rather to offer a **purified and regulated source**
- Clinicaltrials.gov has **six active trials** for Epidiolex and four more completed or unavailable trials, however **none of them have any results posted here**.
- Multiple **news sources report positive results** as described at the committee hearing
- Drugbank does not yet recognize the brand name, but does have some information on cannabidiol
  - A negative allosteric modulator of the CB1 receptor, the most abundant GPCR in the body
  - Activation of 5HT1A serotonergic receptors
  - Antagonizes alpha adrenergic receptors and mu opioid receptors
Epidiolex

From the GW Pharmaceuticals website:

“the exact MOA by which CBD produces its anticonvulsant effects is unknown. Cannabidiol is a structurally novel anti-convulsant. Cannabidiol does not exert its anti-convulsant effects through CB1 receptors, nor through voltage-gated sodium channels. CBD may exert a cumulative anti-convulsant effect, modulating a number of endogenous systems including, but not limited to neuronal inhibition (synaptic and extrasynaptic GABA channels), modulation of intracellular calcium (TRPV, VDAC, GPR55), and possible anti-inflammatory effects (adenosine). CBD does not directly bind to, nor activate, CB1 and CB2 receptors at concentrations pharmacologically relevant to its anticonvulsant effect.”

The label use is for epileptic conditions which are severe and treatment resistant, although the introduction of this product into the market introduces the possibility for off-label prescriptions and increased interest in CBD products.
Studies on CBDs

GW Pharmaceuticals conducted a study to determine if cannabis had any effect on people with epilepsy.

Epidiolex was used during this study. (99% CBD)

- **Results:**
  - 12 week long study with 214 people ranging in age from 2 to 26.
  - All had epilepsy and did not respond well to other forms of treatment.
  - During this study they found that seizures decreased by 54%.
When does 1+1 = 3?

Through the Entourage Effect!
Refers to the potentiating effects of endocannabinoid metabolic byproducts on endocannabinoid function at CB1Rs and CB2Rs

- The sum is greater than the parts
  - The combined effects of compounds found in phytocannabinoids is greater than each compounds effect individually
  - CBD combined with THC, tetrahydrocannabivarin, cannabigerol, cannabichromene, and terpenes
Entourage Effect

- CB receptors, THC, and CBD
  - CBD has a low affinity at CB receptors while THC has a much higher affinity
  - The use of dual pathways may contribute to the mechanism of the entourage effect
- Terpenes
  - Considered safe by FDA
  - B-caryophyllene found in pepper, cinnamon, and other spices
    - Selectively binds to CB2 receptor acting as an agonist
      - Similar to THC, the combined effect of terpenes and CBD could result in greater efficacy
Hormonal Implications in Epilepsy

It has been documented that *increases in estrogen can induce seizures in epileptic patients*.

- This may be because *higher levels of estrogen lead to higher levels of BDNF*
- Increased BDNF in the hippocampus has been shown sufficient to trigger seizure
- Additionally, these same hormonal changes can trigger migraine, a disorder in which BDNF is also abnormally elevated.

**CBD shown to reduce estrogen levels**

- Evidence suggests this is due to the *disruption of hypothalamic GnRH release* and pituitary release of Luteinizing Hormone (LH)
  - Impacts both estrogen and progesterone
- Perhaps one way which CBD may help to manage epilepsy
- A reason why cannabis is being explored in the treatment of breast cancer

A theory: CBD may indirectly repress BDNF, thereby decreasing hormonally-triggered seizure?
Hormones cont.

- **Left diagram shows the (female) HPG axis** Depicts only THC effects on the system
- **CBD exerts inhibitory effects on both the hypothalamus and anterior pituitary**
- **Right diagram shows estrogen’s role in a variety of neural processes**

Scharfman and MacLusky, 2006

Brentz, 2016
CBD is shown to inhibit synaptic transmission in hippocampal slices
- In many forms of epilepsy, seizures originate in the medial temporal lobe, an area that includes the hippocampus. General seizures continue to spread across the entire brain.

Experiment suggests CBD reduces synaptic transmission in hippocampal in vitro through 5HT$_{1A}$ activation and indirect CB1 activation
- Agonist at 5HT$_{1A}$
  - Inhibits Ca$_v$3 subfamily of calcium channels
  - Modulates intracellular calcium levels
- Weak antagonist at CB1
- Weak inverse agonist at CB2
Inhibition cont.

- Cannabidiol reduces spontaneous action potential frequency in cultured hippocampal neurons
  - CBD was without effect on the resting membrane potential at all concentrations tested
  - Effects through Gαi GPCRs
  - Direct activation of 5HT$_{1A}$ receptors + indirect activation of CB1 receptors (via anandamide)
  - 5HT$_{1A}$ receptor antagonism shown to block anxiolytic effects of CBD
(A) CBD reduces firing frequency in dose-dependent manner
(B) CBD inhibited AP frequency more than CB receptor agonists, and effect was diminished by Pertussis toxin, which is a Gαi protein uncoupler.
Anxiolytic Properties

- Stress and lack of sleep are triggers of epileptic seizures
- In rats the elevated plus-maze model is used to trigger anxiety
  - Treatment with **CBD doses of 2.5, 5.0 and 10.0 mg/kg significantly increased the entry ratio** (open/total number of entries)
  - This increase is seen as **CBD working as an anxiolytic**
  - CBD at a dose of 20.0 mg/kg was no longer effective
  - None of the doses of CBD used changed total number of entries instead the ratio to open arms
    - Change in entries would indicate change in exploratory effect
Anxiolytic Properties cont.

Campos and Guimarães, 2008

- $5\text{HT}_{1A}$ is a serotonin autoreceptor located on the cell bodies of serotonergic neurons. Its activation attenuates neural firing.
  - CBD is an agonist at $5\text{HT}_{1A}$
    - Suggested mechanism by which it mediates anxiolytic properties
  - Cannula aimed at dIPAG with CBD solution in elevated plus maze
  - $5\text{HT}_{1A}$ receptor antagonism blocks anxiolytic effects of CBD
  - CB1 antagonism produced no effect on anxiety

Way = $5\text{HT}1A$ antagonist
With CBD exploratory behavior was increased, Way abolished anxiolytic properties
Exploration of CBD in Other Disorders

- TBI
- Migraine
- Bipolar Depression
- Anxiety
- Multiple Sclerosis
- Cancer
- Chronic pain/inflammation
- Diabetes

As time passes, CBD will probably be more thoroughly explored in a number of other disorders with related mechanisms.

On the other hand, cannabis use (though not CBD specifically) has been implicated in schizophrenia and psychotic disorders in at-risk individuals.

- appears more likely that THC is responsible for those effects

A number of these disorders being treated with CBD share genetic markers in common with epilepsy! For example: SCN1A, ATP1A2, CACNA1A: epilepsy, migraine, bipolar
Cannabidiol and Amyloid β Plaques

- Alzheimer's is often accompanied by seizures
- β amyloid derived from the fatty membrane surrounding nerve cells
  - Clump together to form plaques
  - Block cell signaling and result in immune response
- Positive correlation to these plaques and Alzheimer's
- The smaller **oligomers** are theorized to be more harmful due to their ability to move throughout the brain to cause more damage
Cannabidiol and Amyloid β Plaques

- CBD has a high affinity for 5HT\textsubscript{1A} receptors
- These receptors interact through serotonin to inhibit Amyloid precursor protein

- Cannabidiol reduced neuroinflammation in mice injected with Aβ. iNOS and IL-1β expression and release were inhibited
  - Limited conclusions can be made due to the small amount of research done
Drawbacks to CBD

- **Appropriate dosage** is still poorly understood
  - Many different factors to consider (strains, ratios, etc)
- **Many untrustworthy retailers** of the product
  - Especially in states where marijuana is still criminalized
  - 3rd party testing is important
- **Possible drug interactions**
  - Liver enzyme inhibitor, changes the appropriate dose of other drugs
- **Widespread effects**
  - CBD seems to act on many different areas of the brain and body
  - Potential unintended consequences such as decreased fertility
- **Accessibility related to the legality** of cannabis in general
In Summary...

- Current epilepsy medications have many **poor side effects** and don’t always adequately manage epilepsy.
- CBD has **little affinity at CB1 and CB2** receptors, an **agonist at 5HT1A** receptors, and helps **prevent the metabolism and reuptake** of endogenous cannabinoids.
- The **exact mechanisms** by which CBD produces anticonvulsive effects are **unknown**
  - Could be **many different mechanisms**: reducing effects of epileptic triggers and/or changing neuronal excitability.
- Epilepsy has **overlap with many other disorders**, and preliminary results have suggested efficacy of CBD in many of them.
- CBD seems like **novel and effective antiepileptic drug** (AED), especially for poorly controlled, severe, and treatment resistant forms of epilepsy.
Citations

Epilepsy Ontario, epilepsyontario.org/about-epilepsy/types-of-seizures/.
(2003), The History and Stigma of Epilepsy. Epilepsia, 44: 12-14. doi:10.1046/j.1528-1157.44.s.6.2.x
http://www.genecards.org/cgi-bin/carddisp.pl?gene=SCN1A
https://www.drugbank.ca/drugs/DB09061


Citations Cont.


NRSC 4072 Vigers class slides
Thanks :)