CBD/CBDA – rich hemp: Not just for pain anymore!

Dr. Joseph Wakshlag

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CVO –ElleVet Sciences

Professor Cornell University

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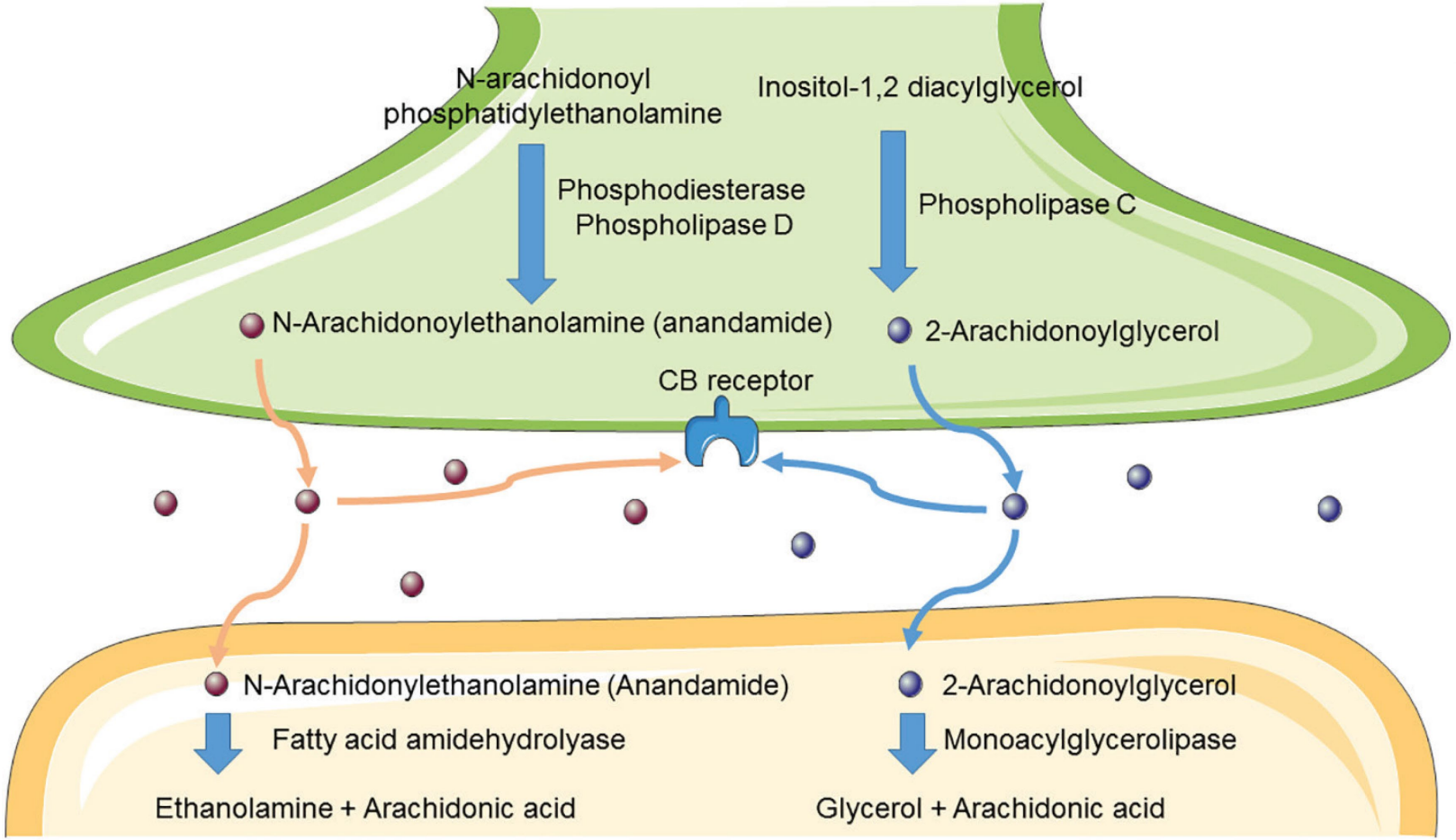
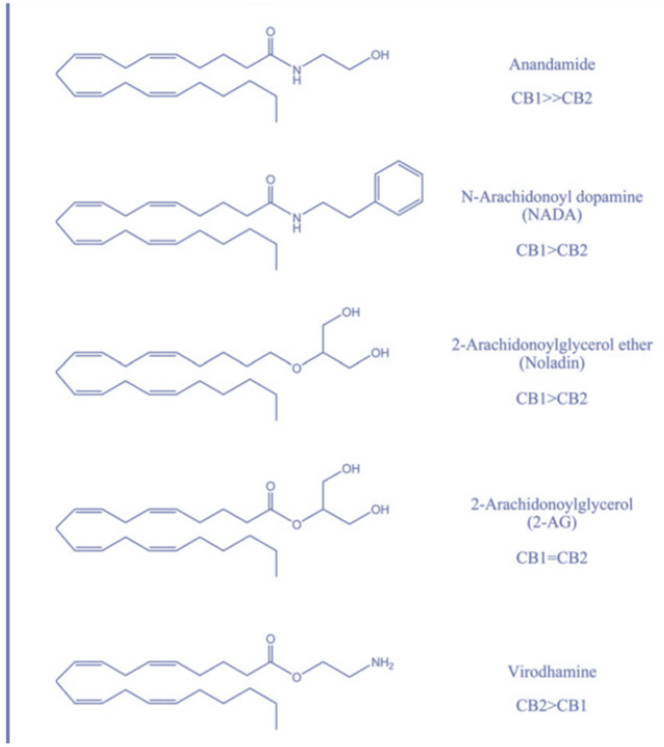
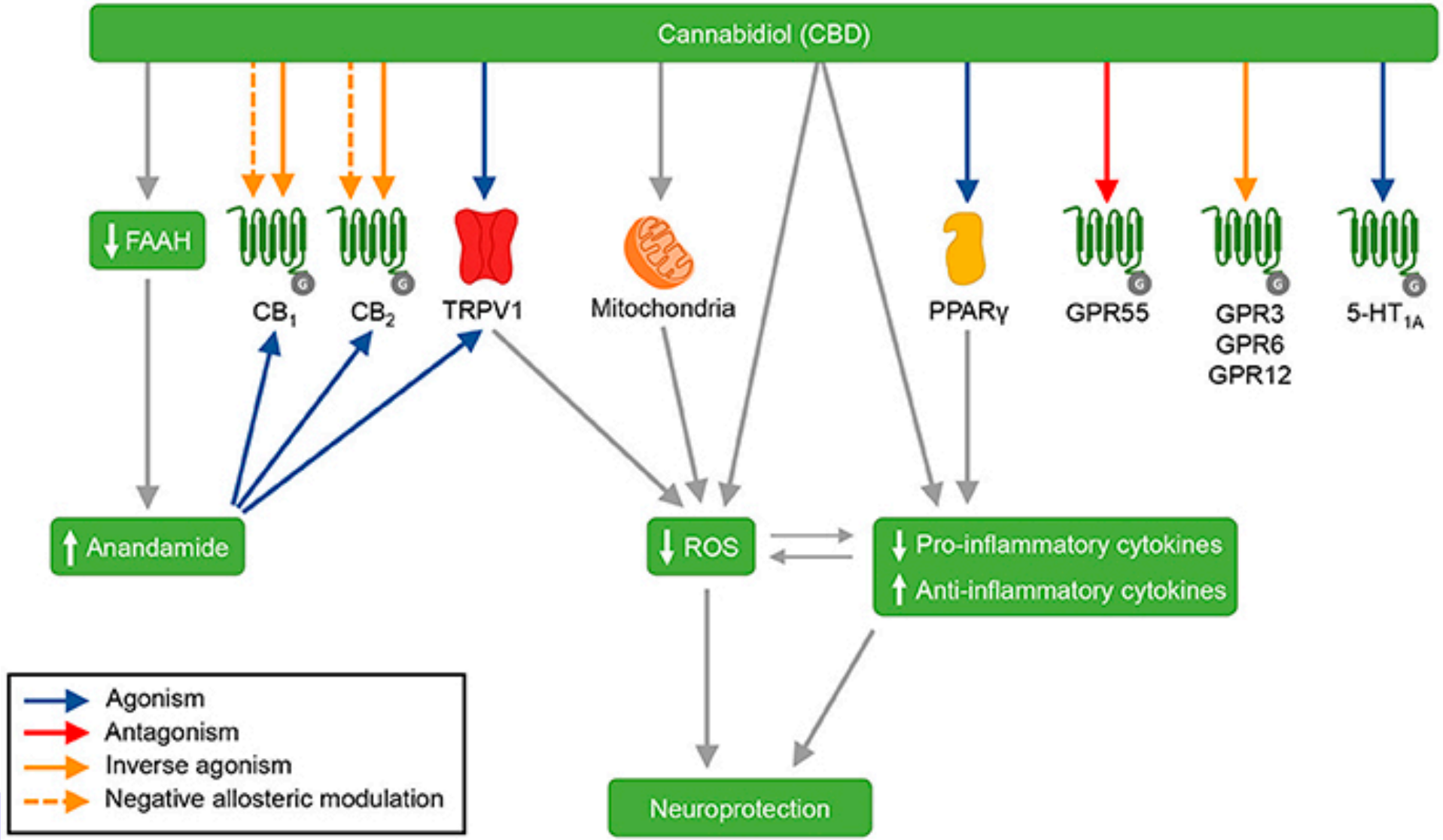
Endocannabinoids and CB Receptors





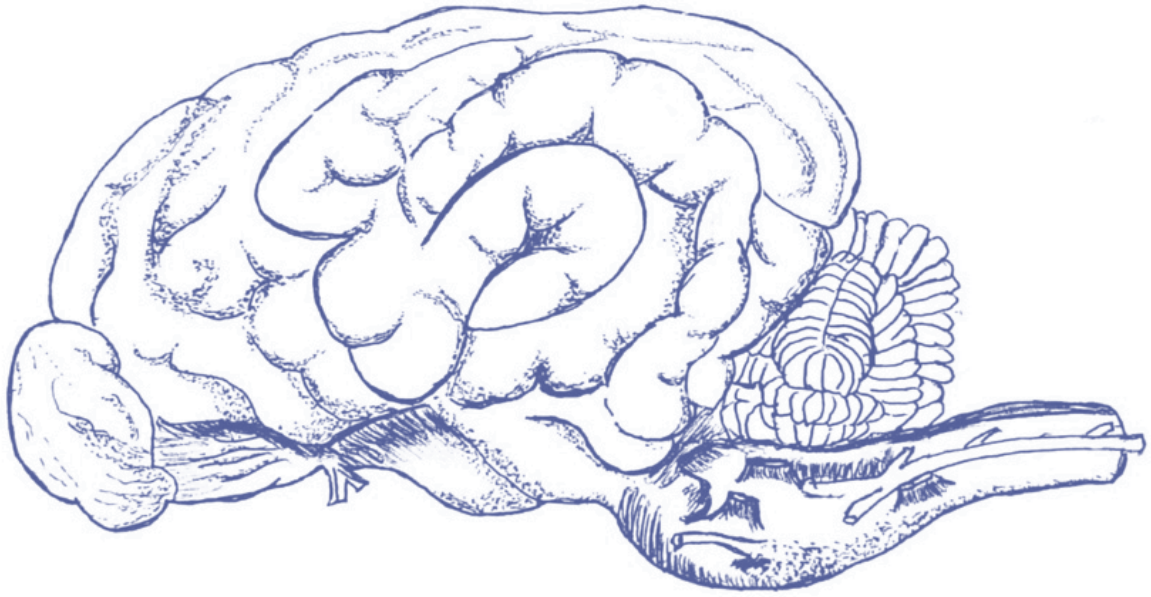
Maayah et al, BAA –molecular basis of dz, 2020

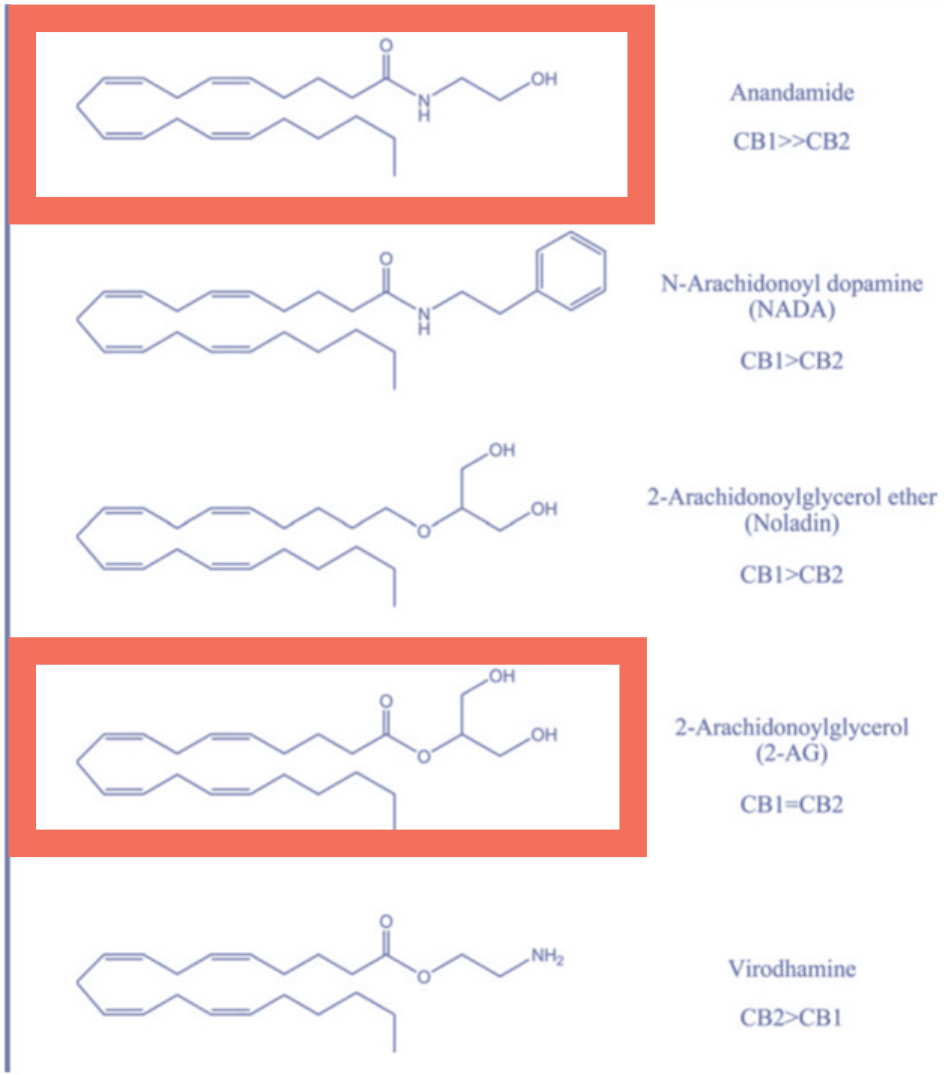
The Endocannabinoidome



Peres, Front in Pharm, 2019

Endogenous Cannabinoids and the Brain – Seizure Control?





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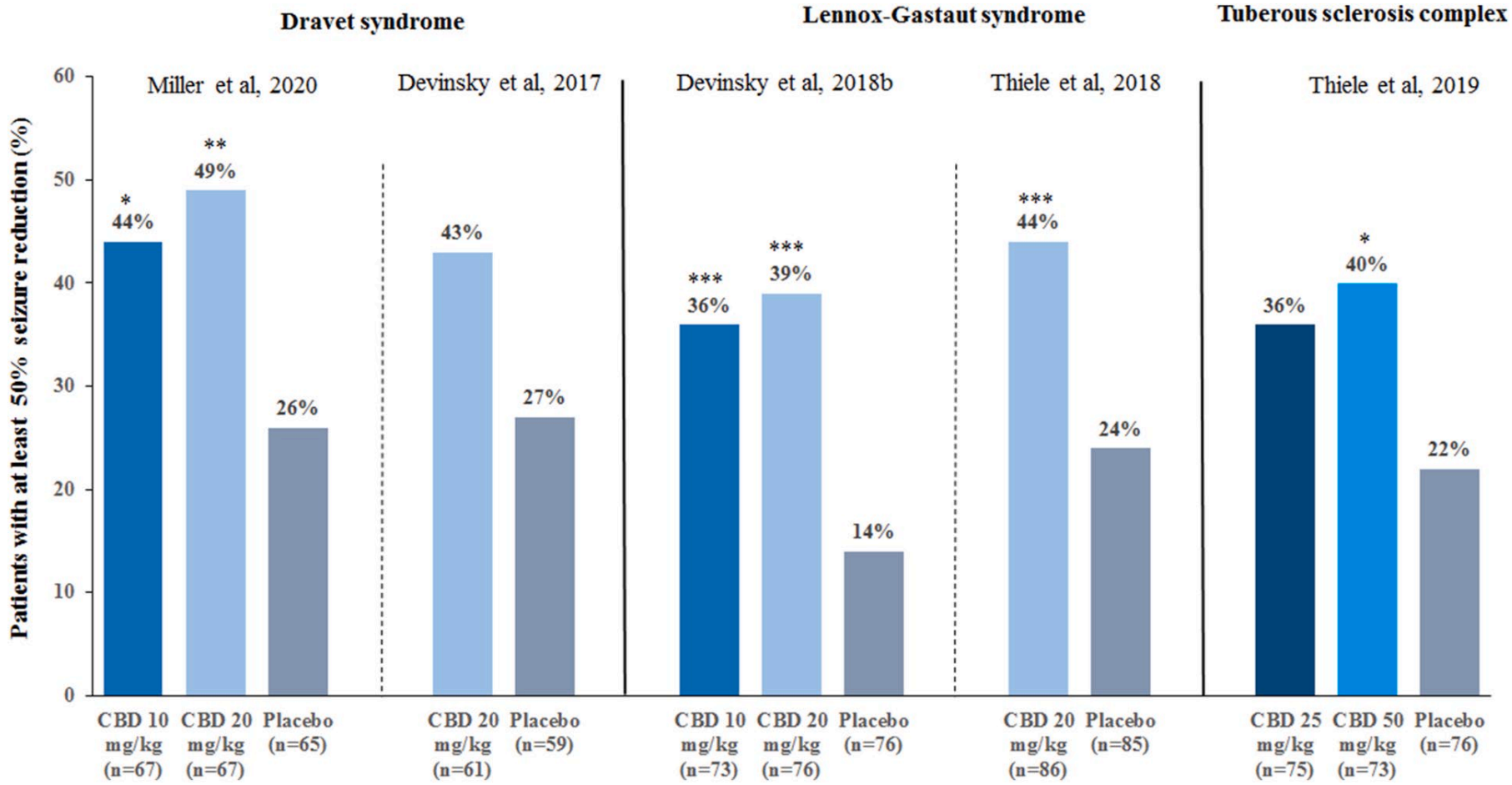
Dravet, Lennox Gastaut, Tuberous Sclerosis Syndromes: The Epidiolex Studies



Franco et al, Neuropharm, 2021

Adverse Events

- Somnolence
- Appetite diminished
- Diarrhea
- Elevated Transaminases

Table 3. Adverse events based on randomised, double-blind, placebo-controlled trials (the most frequent adverse events are highlighted in grey).

	Dravet syndrome				Lennox-Gastaut syndrome			
	Devinsky *et al.*, 2017		Devinsky *et al.*, 2018a		Devinsky *et al.*, 2018b		Thiele *et al.*, 2018	
	CBD (*n*=61)	Placebo (*n*=59)	CBD (*n*=27)	Placebo (*n*=7)	CBD (*n*=149)	Placebo (*n*=76)	CBD (*n*=86)	Placebo (*n*=85)
[CBD] mg/kg/day	20	-	5, 10 or 20-		10 or 20	-	20	-
Reported AEs (%)	93	75	74	86	89	72	86	69
Reported serious AEs (*n*)	10	3	5	1	26	13	20	4
Withdrawn (*n*)	8	1	2	-	7	1	12	1
Somnolence (*n*)*[a]	22	6	5	1	39	4	25	15
Decreased appetite (*n*)	17	3	5	0	32	6	19	3
Pyrexia (*n*)*	9	5	6	0	16	12	12	8
Diarrhoea (*n*)	19	6			19	6	27	10
Elevated transaminases (*n*)*[b]	12	1	6	-	14	0	20	1
Vomiting (*n*)	9	3	3	0	10	9	15	18
Fatigue (*n*)	12	2	1	2				
Upper respiratory infections (*n*)	7	5			21	11		
Pharyngitis (*n*)			3	2	12	5		
Convulsion (*n*)*	7	3	1	2				
Sedation (*n*)			4	0				
Ataxia (*n*)			3	0				
Rash (*n*)*			2	0				
Non-specified pneumonia (*n*)			2	0				
Lethargy (*n*)*	8	3						
Status epilepticus (*n*)	3	3			11	3	1	1

*Serious adverse events reported in ≤2 patients per RCT. [a]Majority of patients were also taking clobazam. [b]>79% patients were taking valproate (transaminases were elevated >3 times the upper normal limit).

Arzimanoglou et al, Epileptic Disorders, 2020

CSU's Epilepsy Study

Randomized blinded controlled clinical trial to assess the effect of oral cannabidiol administration in addition to conventional antiepileptic treatment on seizure frequency in dogs with intractable idiopathic epilepsy

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OBJECTIVE
To assess the effect of oral cannabidiol (CBD) administration in addition to conventional antiepileptic treatment on seizure frequency in dogs with idiopathic epilepsy.

DESIGN
Randomized blinded controlled clinical trial.

ANIMALS
26 client-owned dogs with intractable idiopathic epilepsy.

PROCEDURES
Dogs were randomly assigned to a CBD (n = 12) or placebo (14) group. The CBD group received CBD-infused oil (2.5 mg/kg [1.1 mg/lb], PO) twice daily for 12 weeks in addition to existing antiepileptic treatments, and the placebo group received noninfused oil under the same conditions. Seizure activity, adverse effects, and plasma CBD concentrations were compared between groups.

RESULTS
2 dogs in the CBD group developed ataxia and were withdrawn from the study. After other exclusions, 9 dogs in the CBD group and 7 in the placebo group were included in the analysis. Dogs in the CBD group had a significant (median change, 33%) reduction in seizure frequency, compared with the placebo group. However, the proportion of dogs considered responders to treatment (≥ 50% decrease in seizure activity) was similar between groups. Plasma CBD concentrations were correlated with reduction in seizure frequency. Dogs in the CBD group had a significant increase in serum alkaline phosphatase activity. No adverse behavioral effects were reported by owners.

CONCLUSIONS AND CLINICAL RELEVANCE
Although a significant reduction in seizure frequency was achieved for dogs in the CBD group, the proportion of responders was similar between

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CSU's Epilepsy Study:

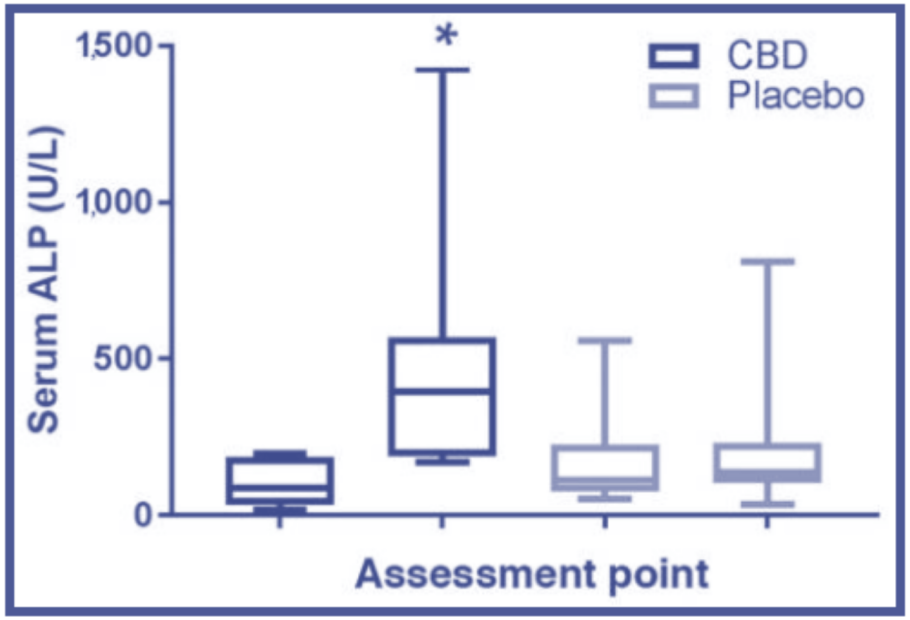

Figure 1—Box-and-whisker plots of serum ALP activity at week 0 (before study treatment) and week 12 for client-owned dogs with intractable idiopathic epilepsy that were randomly assigned to receive CBD-infused oil (2.5 mg/kg [1.1 mg/lb], PO, twice daily for 12 weeks; n = 9; black boxes) or a placebo at a similar dosage (7; gray boxes), in addition to currently prescribed conventional AEDs. The top and bottom of each box represent the 75th and 25th percentiles, respectively; the central horizontal line within each box represents the median; and the whiskers represent the minimum and maximum values. *Values differ significantly (P = 0.004) between assessment points for dogs in the CBD group.

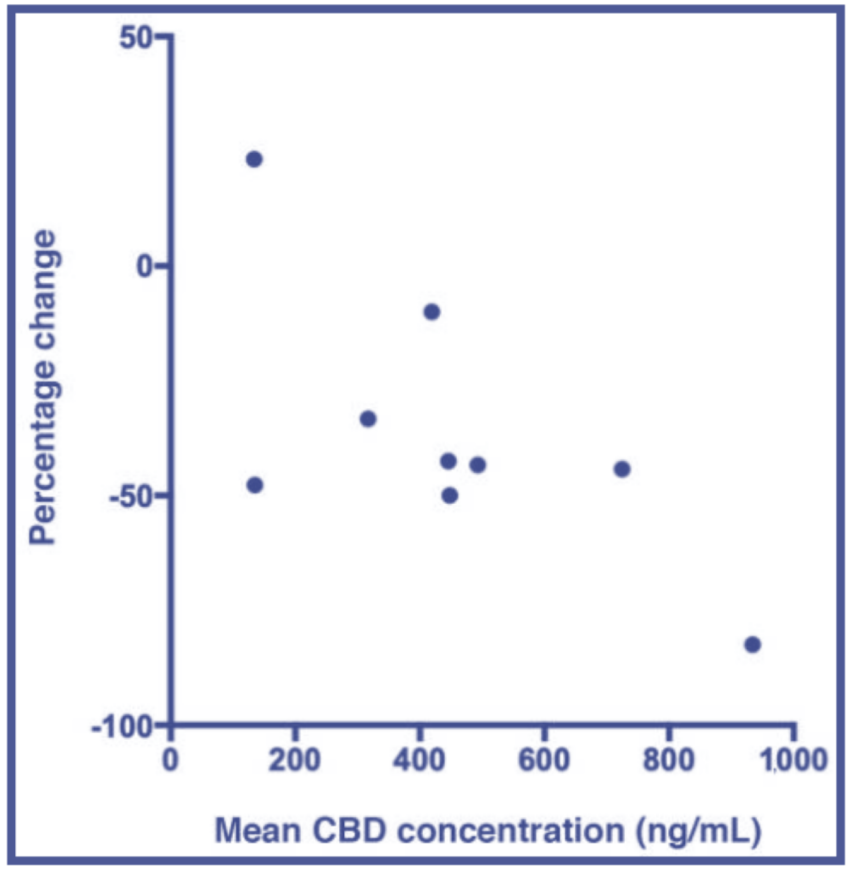

Figure 2—Dot plot showing the negative correlation (r = –0.68; P = 0.04) between mean plasma CBD concentration (measured at weeks 4, 8, and 12) and the percentage change from before study treatment began (week 0) in mean monthly seizure frequency for dogs in the CBD group (n = 9).

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Cannabidiol As An Adjunct In Drug-Resistant Epilepsy • Rozenthal et al. (2023)

- Randomized, double blinded, placebo-controlled crossover trial
- ≧ 2 seizures/month for at least 12 weeks
- At least one of phenobarbital, potassium bromide, levetiracetam and zonisamide
- 61 dogs initially enrolled to trial
- Chicken flavored CBD-infused hemp seed oil **OR** chicken flavored hemp seed oil as placebo
- 4 week washout between treatments
- No ASM changes allowed, although rescue dosing for 3-5 days was permitted
  - → Levetiracetam, gabapentin, clorazepate or combination

5mg/kg/day (2.5mg/kg BID) dose used initially for 13 dogs

→ Lack of efficacy during treatment, so dose changed

Remaining 48 dogs administered **9mg/kg/day (4.5mg/kg BID)** in treatment phase

Rozental AJ, J Vet Intern Med. 2023 Nov-Dec;37(6):2291-2300.

Cannabidiol As An Adjunct In Drug-Resistant Epilepsy • Rozenthal et al. (2023)

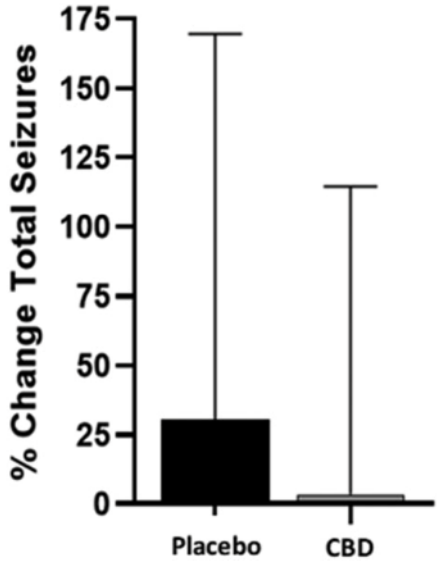

FIGURE 1 Means and SD of percent change of total seizures from baseline averaged over the course of 3 months while taking 9 mg/kg/day of CBD oil versus placebo, along with conventional ASD treatment ($n = 39$). Treatment $P = .04$. CBD, cannabidiol.

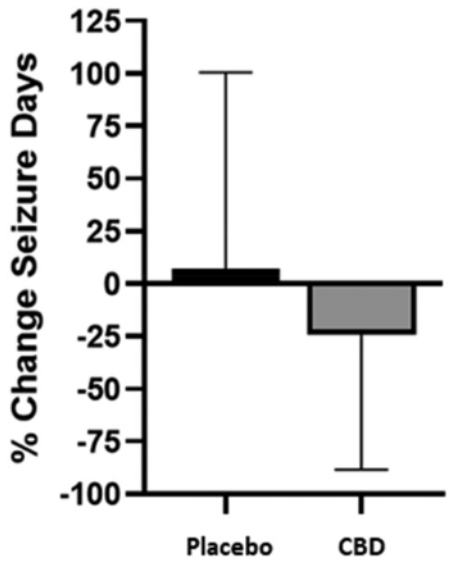

FIGURE 2 Means and SD of percent change of seizure days from baseline averaged over the course of 3 months while taking 9 mg/kg/day of CBD oil versus placebo, along with conventional ASD treatment ($n = 39$). Treatment $P = .002$. CBD, cannabidiol.

Cannabidiol As An Adjunct In Drug-Resistant Epilepsy • Rozenthal et al. (2023)

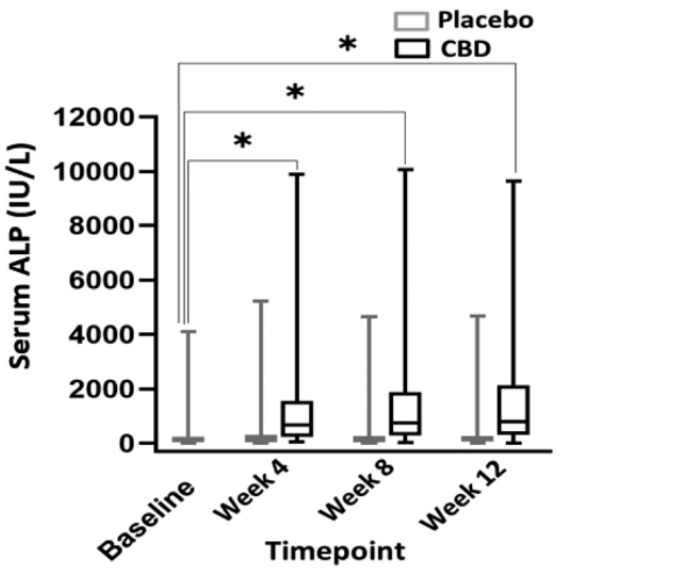

FIGURE 3 Box and whisker plot comparing serum ALP activity from baseline over the course of 3 months while taking 9.0 mg/kg/day of CBD oil versus placebo, along with conventional ASD treatment ($n = 39$). There was a significant difference between CBD treatment and baseline in all months (asterisks) ($P \leq .0001$). There was no difference between placebo and baseline in any month ($P \geq .88$). ALP, alkaline phosphatase; CBD, cannabidiol.

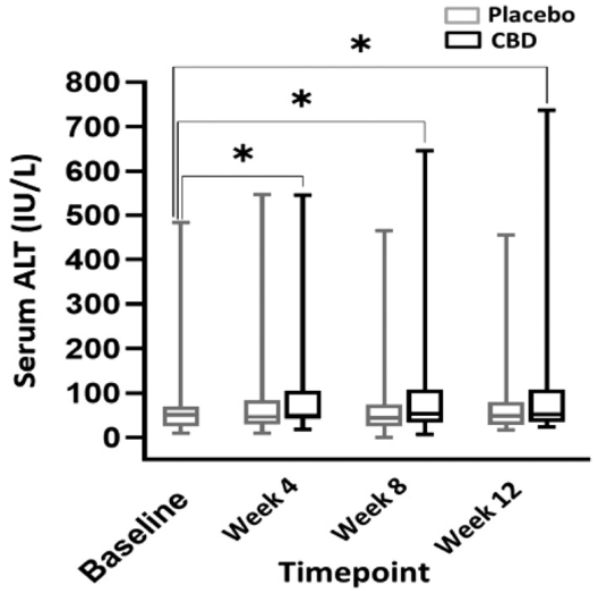

FIGURE 4 Box and whisker plot comparing serum ALT activity from baseline over the course of 3 months while taking 9 mg/kg/day of CBD oil versus placebo, along with conventional ASD treatment ($N = 39$). There was a statistically significant increase in ALT with CBD treatment when compared to baseline (asterisks) ($P \leq .003$). There was no difference between placebo and changes in ALT. ALT, alanine transaminase; CBD, cannabidiol.

Also showed no alterations in Phenobarbital or bromide during treatment!

ElleVet Seizure Study: Methods

- Two Centers participated
  - University of Florida –Dr. Gabriel Garcia
  - Vet. Neuro. and Pain Mngmt Center of NE
    - Dr. Stephanie Kube
- Placebo Blinded Cross Over Design
  - Dogs with at least monthly grand mal seizures.
  - Refractory to common medications
  - Can be on zonisamide, keppra, phenobarbital, potassium bromide.
  - 10 of 14 dogs enrolled have MRI and CSF tap negative for infectious or structural
  - 4 dogs refractory multiple year treatment – refractory epilepsy based on history.
- Randomized into treatment groups
  - Treatment phase – 2 mg/kg of a full spectrum hemp containing 50: 50 mix of CBD:CBDA in sesame seed oil filled capsules
  - THC less than 0.3% with ratio of CBD/CBDA to THC/THCA being 27:1
  - Placebo Group – sesame seed oil
  - All Placebo jars liked with hemp oil to provide distinctive smell.
- 14 dogs have finished with data for examination
- Paper published this past Summer!

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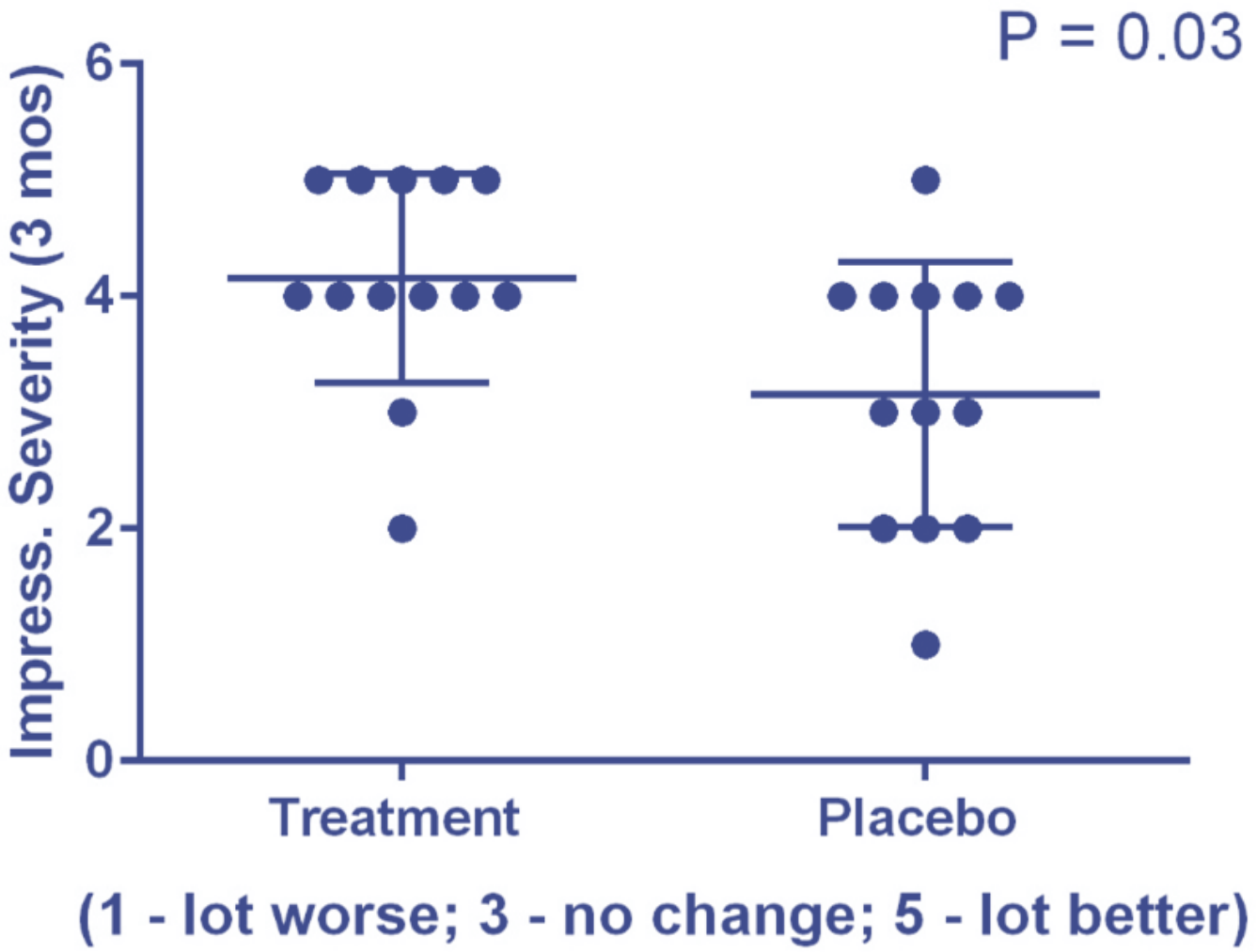
ElleVet Results: Owner Survey



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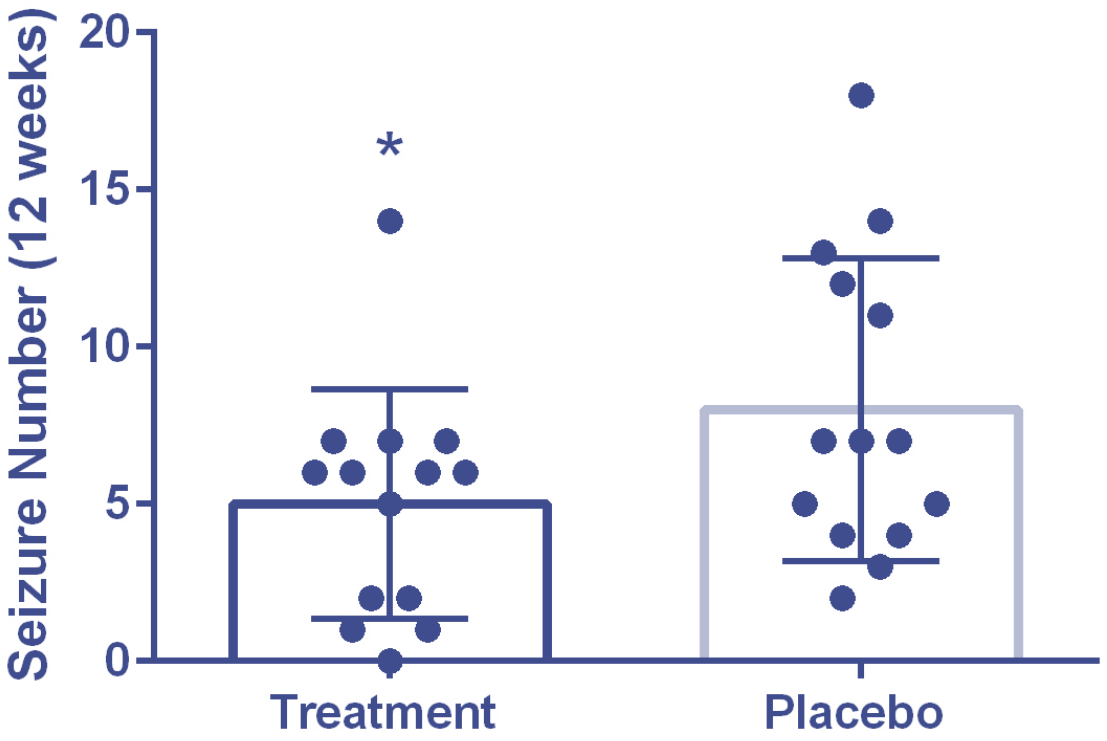
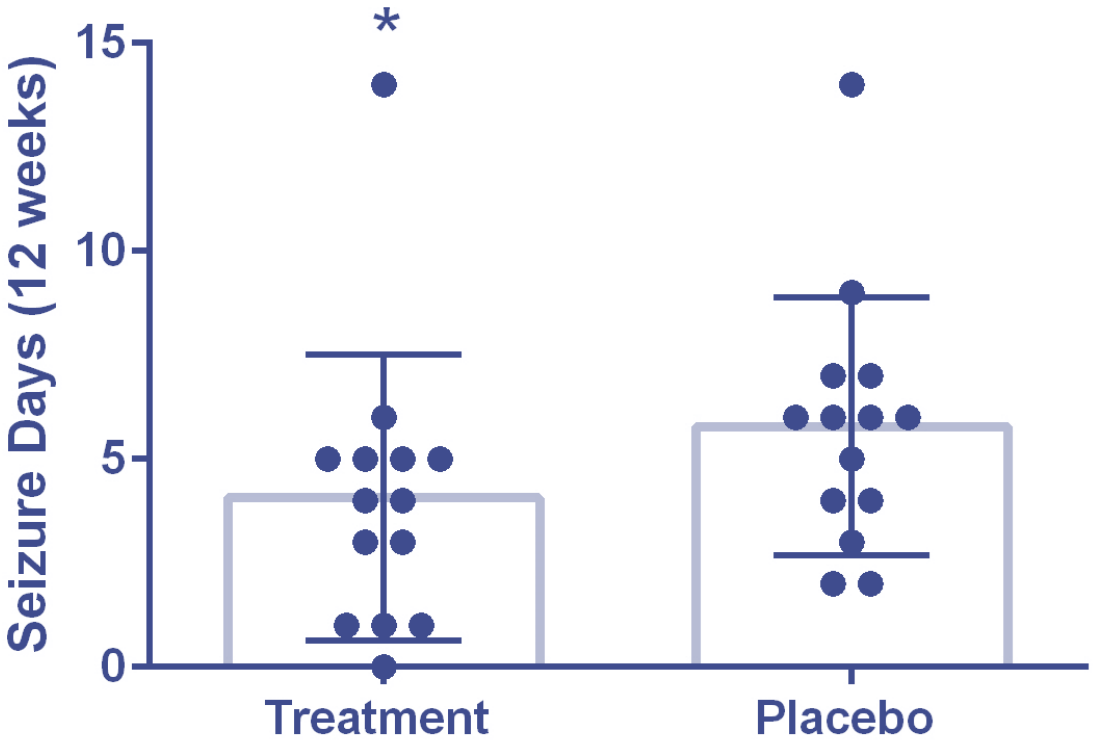
ElleVet Results: Seizure Diary





6 dogs during Ellevet treatment has a 50% decrease in frequency or days – none during placebo treatment

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ElleVet Results: Adverse Events

- Owners Surveyed at 3 months
- Owners asked about increased hunger, vomiting, diarrhea, weight gain, thirst, urination, sleepiness, restlessness/anxiety, ataxia.
- Scores of 1-3 (no change or decrease vs 4 or 5 (increased)
- Fisher's Exact test performed

Adverse Events 13 respondents	Placebo	Treatment	Fisher's Exact
Increased appetite	0	3	p = 0.22
Increased GI signs(vomit/diarrhea)	0	2	p = 0. 48
Increased thirst/urination	3	2	p = 1.0
Increased Ataxia	1	4	p = 0.32
Increased Anxiety	3	2	p = 1.0
Increased lethargy/somnolence	1	3	p = 0.59

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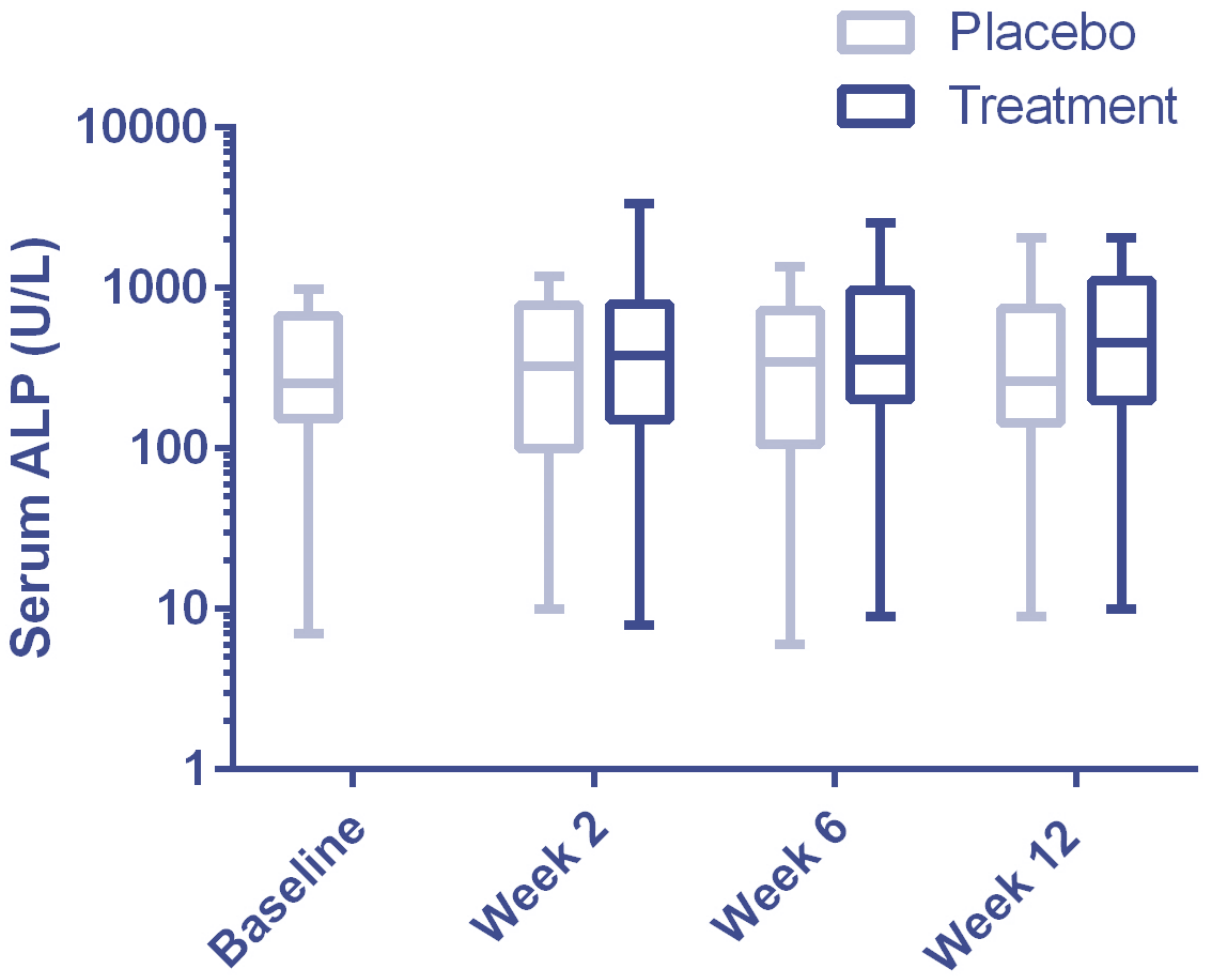
ElleVet Results: Serum Chemistry



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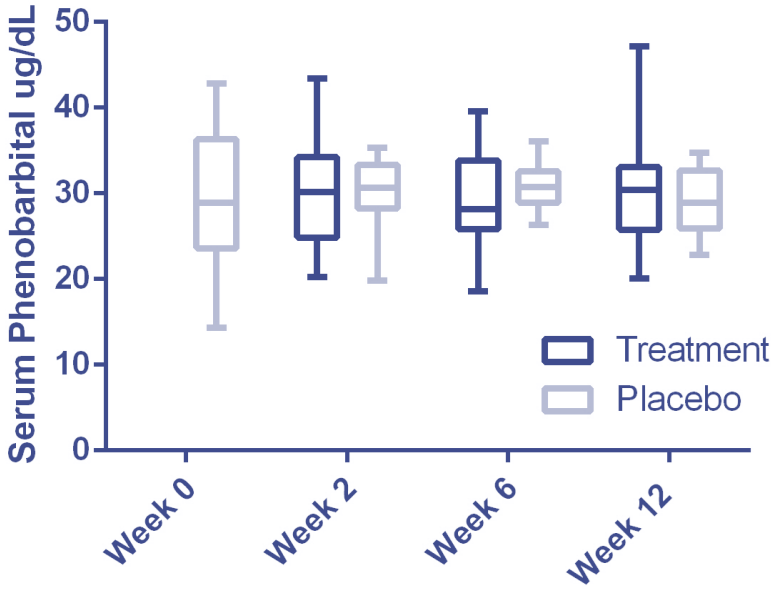
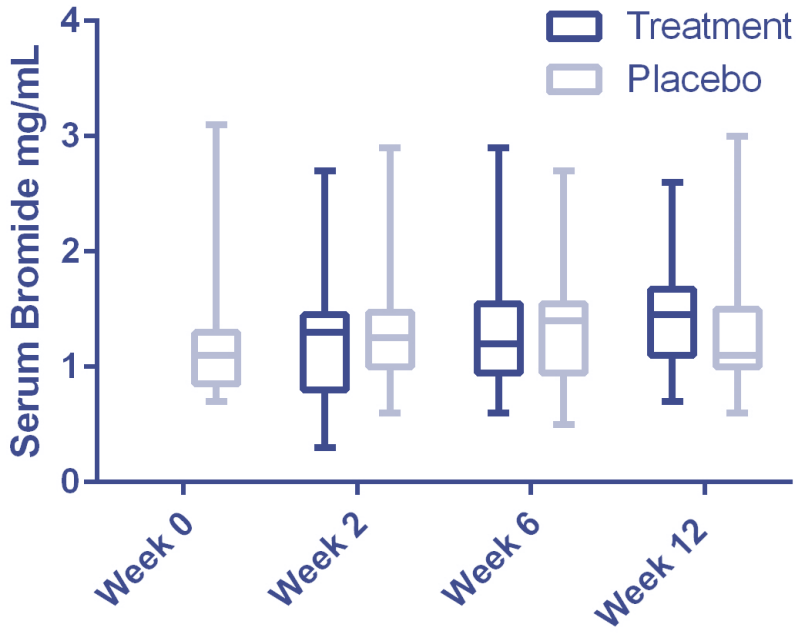
ElleVet Results: AED's

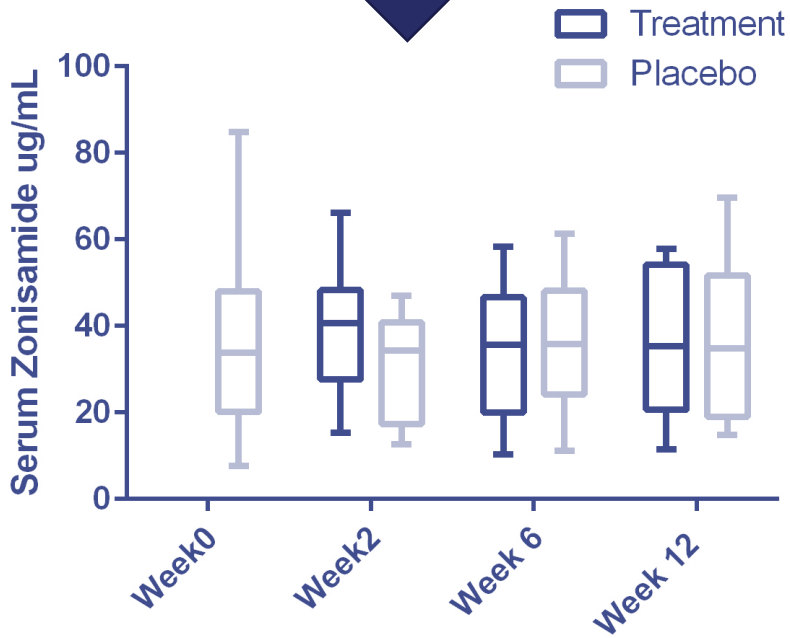
Phenobarbital (n = 9)

KBr (n = 13)

Zonisamide (n = 11)







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Recent Seizure Work:

frontiers in Neurology

BRIEF RESEARCH REPORT
published: 03 July 2019
doi: 10.3389/fneur.2019.00716

Dosage Related Efficacy and Tolerability of Cannabidiol in Children With Treatment-Resistant Epileptic Encephalopathy: Preliminary Results of the CARE-E Study

Richard J. Huntsman[1,2], Richard Tang-Wai[1,3], Jane Alcorn[1,4], Stephanie Vuong[4], Bryan Acton[1,5], Scott Corley[1,6], Robert Laprairie[1,4], Andrew W. Lyon[1,7], Simona Meier[6], Darrell D. Mousseau[1,8], Doris Newmeyer[2], Erin Prosser-Loose[2], Blair Seifert[1,9], Jose Tellez-Zenteno[1,10], Linda Huh[11], Edward Leung[12] and Philippe Major[13]*

Conclusion: The preliminary data suggest an initial CBD target dose of 5–6 mg/kg/day when a 1:20 THC:CBD CHE is used. Possible non-linear pharmacokinetics of CBD and CBC needs investigation. The reduction in seizure frequency seen suggests improved seizure control when a whole plant CHE is used. Plasma THC levels suggest a low risk of THC intoxication when a 1:20 THC:CBD CHE is used in doses up to 12 mg/kg CBD/kg/day.

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Recent Seizure Work:

frontiers in Neurology

SYSTEMATIC REVIEW
published: 12 September 2018
doi: 10.3389/fneur.2018.00759

Potential Clinical Benefits of CBD-Rich *Cannabis* Extracts Over Purified CBD in Treatment-Resistant Epilepsy: Observational Data Meta-analysis

Fabricio A. Pamplona[1], Lorenzo Rolim da Silva[2] and Ana Carolina Coan[3]*

(94/223, 42%). Patients treated with CBD-rich extracts reported lower average dose (6.0 mg/kg/day) than those using purified CBD (25.3 mg/kg/day). The reports of mild (158/216, 76% vs. 148/447, 33%, $p < 0.001$) and severe (41/155, 26% vs. 23/328, 7%, $p < 0.0001$) adverse effects were more frequent in products containing purified CBD than in CBD-rich extracts. CBD-rich extracts seem to present a better therapeutic profile than purified CBD, at least in this population of patients with refractory epilepsy. The roots of this difference is likely due to synergistic effects of CBD with other phytocompounds (aka Entourage effect), but this remains to be confirmed in controlled clinical studies.

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Anxiety and People and Dogs!

Contents lists available at ScienceDirect

Journal of the American Pharmacists Association

journal homepage: www.japha.org

REVIEW

Use of cannabidiol in anxiety and anxiety-related disorders

Jessica W. Skelley*, Crystal M. Deas, Zachary Curren, Jonathan Ennis

- Theory behind use is CBD ability to decrease natural anandamide metabolism increasing cannabinoid tone
- Activation of the 5HT1 receptor

The Impact of Feeding Cannabidiol (CBD) Containing Treats on Canine Response to a Noise-Induced Fear Response Test

*Elizabeth M. Morris[1], Susanna E. Kitts-Morgan[2], Dawn M. Spangler[2], Kyle R. McLeod[1], Joao H. C. Costa[1] and David L. Harmon[1]**

TABLE 3 | Ethogram of behaviors tracked by a single trained observer blinded to treatments using The Observer XT (Noldus Information Technology Inc., Leesburg, VA).

Behavioral category	Behavior	Definition used
Movement	Inactive	Standing still, sitting, or laying down
	Cowering	Sudden cessation of movement in response to a stimulus
	Pacing	Frantically moving back and forth, restlessness
	Destruction	Scratching or chewing at room furnishings
Eyes	Facing door	Eyes are focused on the door of the room
	Glancing around	Eyes are shifting back and forth, possibly looking for the source of a sound
	Other	Eyes are focused on something else in the room
Ears	Ears relaxed	Ears are held in natural position
	Ears erect	Ears raised in response to stimulus
	Ears moving	Ears moving back and forth
Tail posture	Tail relaxed	Tail is not rigid and is lower than the top of the body
	Tail stiff	Tail is rigid and horizontal
	Tail wagging	Tail is wagging back and forth
	Tail tucked	Tail is tucked between hind legs
Muzzle	Barking	Emitting a short, loud sound
	Whining	Emitting a long, high pitch sound, often repeated
	Panting	Mouth open wide with tongue protruding while breathing heavily
	Licking	Using the tongue on own body or another object
	Yawning	Opening the mouth wide and inhaling
	Biting	Using teeth on the door or object

CBD Anxiety in Dogs

- 24 dogs recruited from shelter after screening – 16 finished study.
- Housed at LMU in one of two kennels during stay and fed a standard diet.
- Given control treat, trazodone, CBD treat (1.4 mg/kg ave) or both.
- 3 days of room acclimation for 6 minutes
- Then fireworks testing – 3 minutes no noise and then 3 minutes of fireworks.
- Dogs rotated randomly through protocol treatment each week with daily acclimation to the room
- CBD given 4-6 hrs before test; trazadone given 2-4 hrs before test.
- Blood collected before, immediately after and 1 hr after for cortisol and CBD analysis.

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Results:

- Heart Rate mildly higher in CBD group (p = 0.09)
  – Interesting since THC is tachy and CBD is not.
- Serum cortisol lower is lower with trazadone from baseline
  – not CBD or both together.

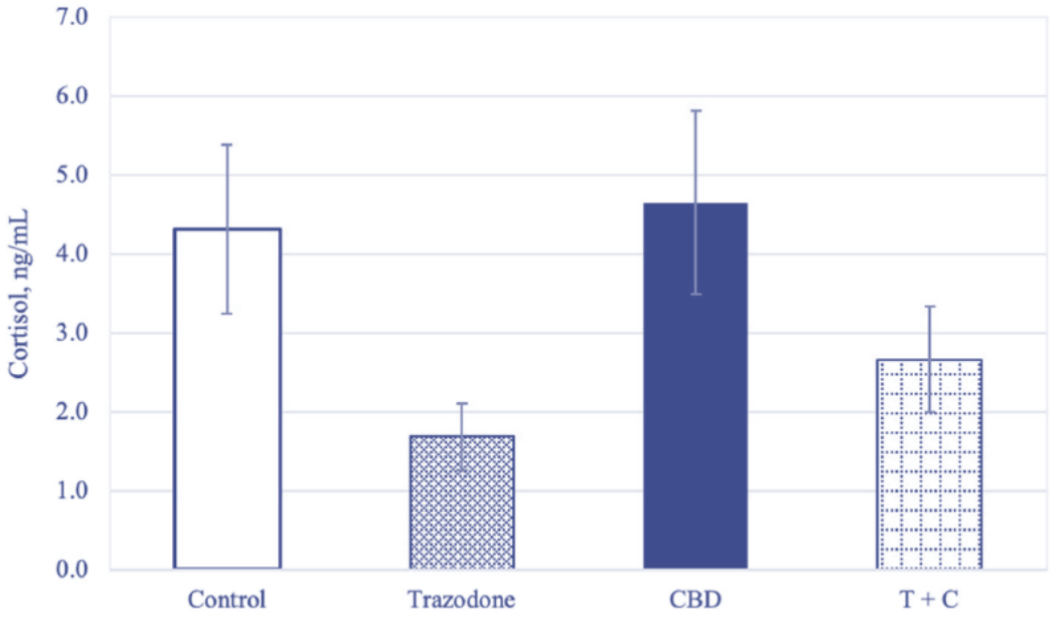

FIGURE 1 | Cortisol concentration (ng/mL) for each treatment ($n = 16$), back transformed after analysis. Error bars represent the standard error of the treatment mean (SEM), which was calculated from the back-transformed confidence interval for each treatment: SEM = (upper limit—lower limit)/3.92. Due to lack of effect of time ($P = 0.189$) and any interactions with time ($P > 0.05$), all time points (Pre-Noise and Noise) have been combined. Trazodone treatment reduced cortisol concentration ($P < 0.001$), whereas there was no effect of CBD ($P = 0.104$) nor the CBD by trazodone interaction ($P = 0.238$).

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Anxiety Behavioral outcomes

TABLE 5 | Effect of trazodone (T), CBD (C), CBD by trazodone interaction (C*T), time (Pre-Noise and Noise), CBD by trazodone by time interaction (C*T*Time), and period on the duration of behavioral parameters (s) for 1-min immediately prior to (Pre-Noise) and the first minute (Noise) of the noise-induced fear response tests administered after each 7-d treatment period.

Parameter, s	Treatment				SE[1]	*P*-value					
	Control	Trazodone (T)	CBD (C)	T+C		Trazodone	CBD	C*T	Time	C*T*Time	Period
Inactive	55.35	56.33	55.21	56.26	1.214	0.329	0.918	0.971	0.011	0.092	0.993
Facing door	37.45	33.90	34.96	37.70	4.198	0.872	0.796	0.217	0.561	0.556	0.786
Glancing around	16.90	15.65	17.93	15.91	3.460	0.396	0.736	0.841	<.001	0.142	0.819
Other eyes	5.10	13.33	4.10	5.48	1.885	0.044	0.072	0.182	<.001	0.469	0.792
Ears relaxed	11.37	7.76	12.35	11.43	4.913	0.179	0.168	0.422	<.001	0.868	0.567
Ears erect	29.33	34.29	29.93	29.80	5.614	0.304	0.408	0.279	<.001	0.747	0.982
Ears moving	19.25	17.79	17.20	18.78	2.076	0.070	0.742	0.351	<.001	0.457	0.493
Tail relaxed	37.90	49.86	38.93	50.96	4.857	0.001	0.753	0.992	0.611	0.898	0.990
Tail stiff	18.45	5.55	16.39	6.65	4.582	0.002	0.887	0.644	0.010	0.757	0.896

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OPEN

Cannabis sativa L. may reduce aggressive behaviour towards humans in shelter dogs

Sara Corsetti[1]✉, Simona Borruso[2], Livia Malandrucco[3], Valentina Spallucci[4], Laura Maragliano[3], Raffaella Perino[3], Pietro D'Agostino[5] & Eugenia Natoli[3]

- Shelter situation with behavioral scoring of aggressive tendancies towards humans
- 12 treatment dogs 12 placebo treatment dogs
  - Obscure treatment of 1 drop per 2 kg of a "CBD olive oil"
  - 150 mg/L of cannabinoids
  - 150 mg/ml assuming a drop between 0.05-1 ml – somewhere between 3.5-7.5 mg/kg assumption
- Dogs acclimated to observation by 2 blinded observers – behavioral experts.
- Observed 1 hr three times a day for on each study day for aggressive behaviors, stereotypic stress behaviors and mentation environmental interactions
- Baseline – 15 days, 45 days and 15 days after stopping administration.

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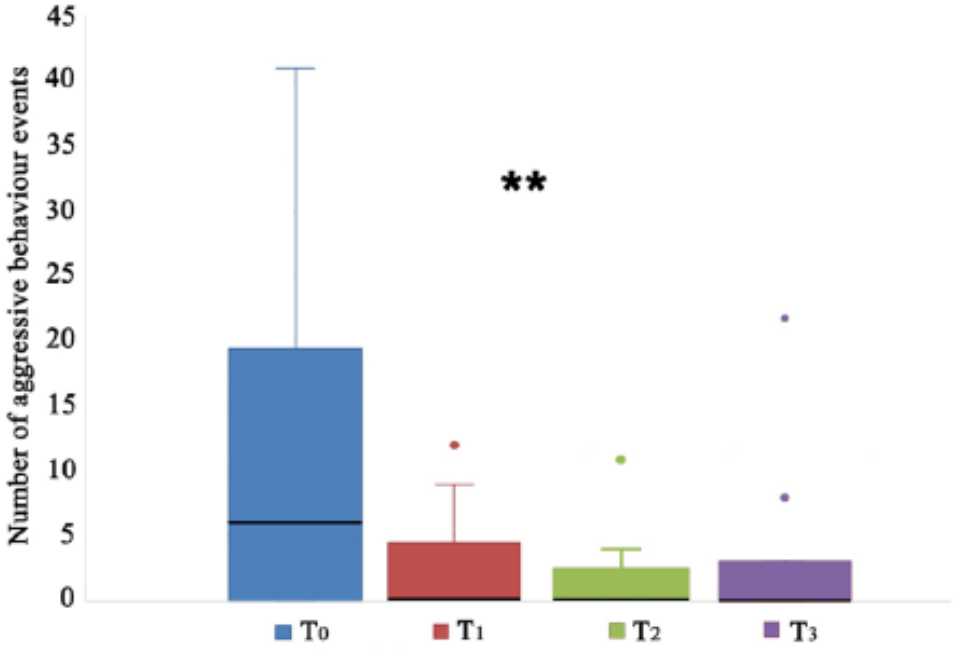
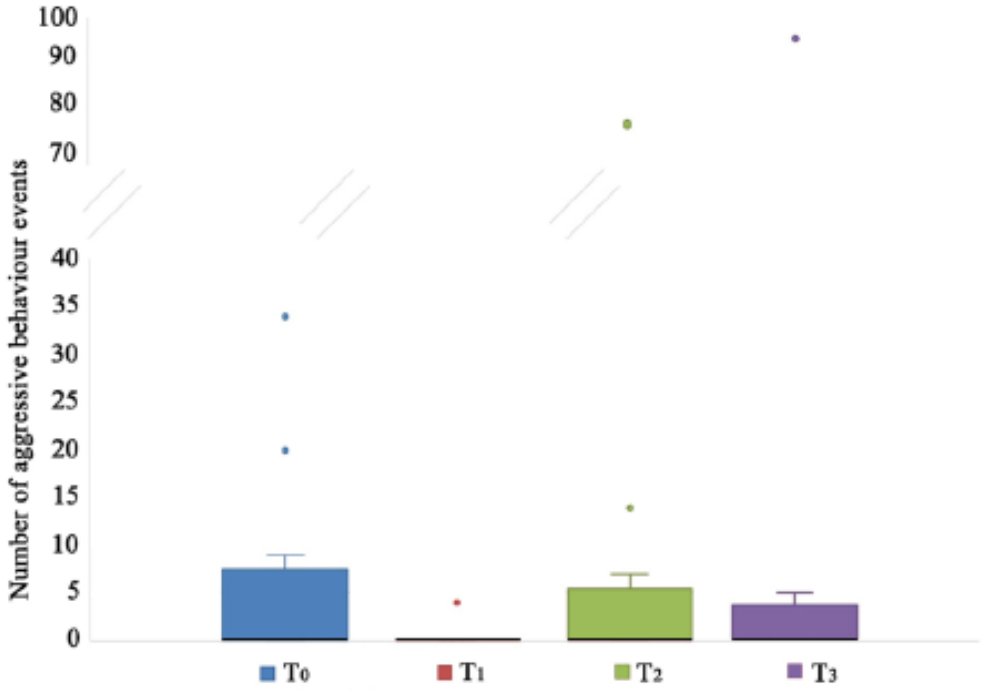
Aggressive behaviors:





Stereotypic kenneled behaviors and mentation/environment interactions were not different between groups or over time in either placebo or treatment groups

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Thoughts:

- Is 1.4 mg/kg PO enough?
- Is the timing good when we know that peak is 1-2 hrs and most is gone by 6 hrs.
- How severe was the firework phobia in shelter dogs? Not client owned.
- How good is CBD at activating the 5HT1 receptor? Maybe there is something better?

British Journal of Pharmacology

British Journal of Pharmacology (2018) **175** 100–112 100

RESEARCH PAPER

Cannabidiolic acid methyl ester, a stable synthetic analogue of cannabidiolic acid, can produce 5-HT_{1A} receptor-mediated suppression of nausea and anxiety in rats

Psychopharmacology (2017) 234:2207–2217
DOI 10.1007/s00213-017-4626-5

ORIGINAL INVESTIGATION

Effect of prior foot shock stress and Δ^9-tetrahydrocannabinol, cannabidiolic acid, and cannabidiol on anxiety-like responding in the light-dark emergence test in rats

Erin M. Rock[1] · Cheryl L. Limebeer[1] · Gavin N. Petrie[1] · Lauren A. Williams[1] · Raphael Mechoulam[2] · Linda A. Parker[1]

Cannabidiol disrupts conditioned fear expression and cannabidiolic acid reduces trauma-induced anxiety-related behaviour in mice

Neda Assareh[a,b,c], Anand Gururajan[a,b,d], Cilla Zhou[a,b,e], Jia Lin Luo[a,b,d], Richard C. Kevin[a,b,d] and Jonathon C. Arnold[a,b,e]

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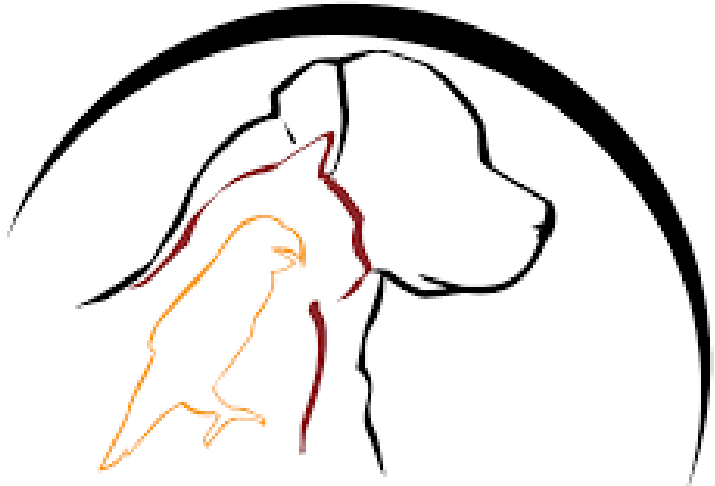
Thunderstorm Phobia situational use study:

Dr. Lisa Radosta

Study Storm Phobia in Dogs



- Design (15 dogs)
- Initial Screen of dogs for Thunderstorm Phobia for eligibility
- Dispense a higher dose chew at approximately 4 mg/kg – Ellevet Calm and Comfort Chew vs Placebo
- Dogs provided chew 1 hr before recorded Thunderstorm (same decibals) with video at office.
- Validated Owner scored behaviors immediately after and 15 minute after event
- Behaviorist video assessment of 10 second interval of anxiety and fear behavior or postures.

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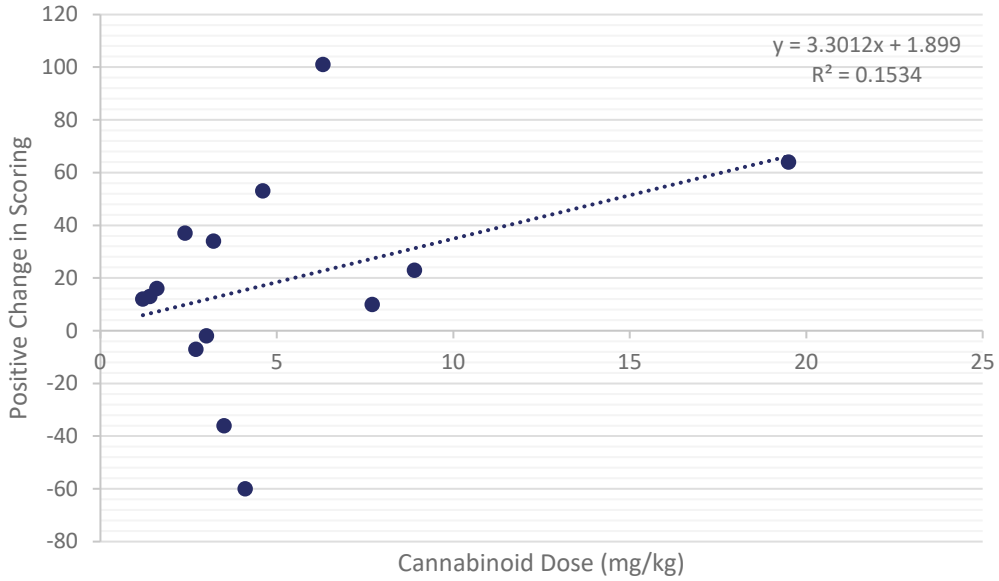
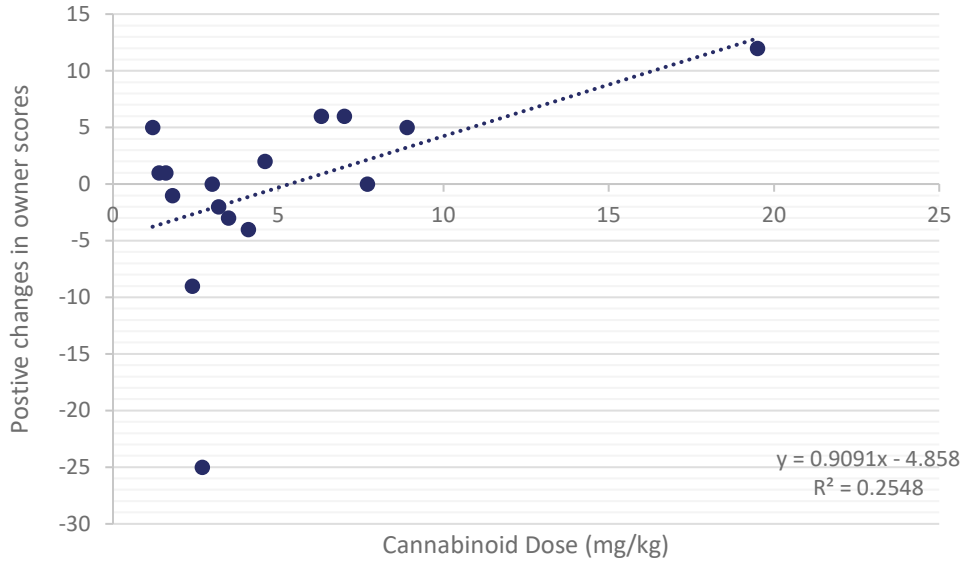
Thunderstorm Phobia situational use:

- All best plans can sometimes be laid in waste – owners provided only 1 chew regardless of weight of dog providing a range of 1.5-19 mg/kg given.





We still need to find the right dose for CBD/CBDA use for fear/anxiety conditions. And one dose may not fit all

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Cancog Car Ride Model

- 10 dogs in each group
  - Placebo soft gel
  - 4 mg/kg Ellevet
  - 6 mg/kg Ellevet
- 10 minute car ride model on week one without treatments
- Week later repeated with treatments 1.5 hrs after given treatment
  - Serum cortisol
  - Heart Rate Variability
  - Video assessment – salivation, lip licking, sitting/standing/lying, vocalization, cage escape.
- Goal to assess apples to apples – car ride 1 vs car ride 2

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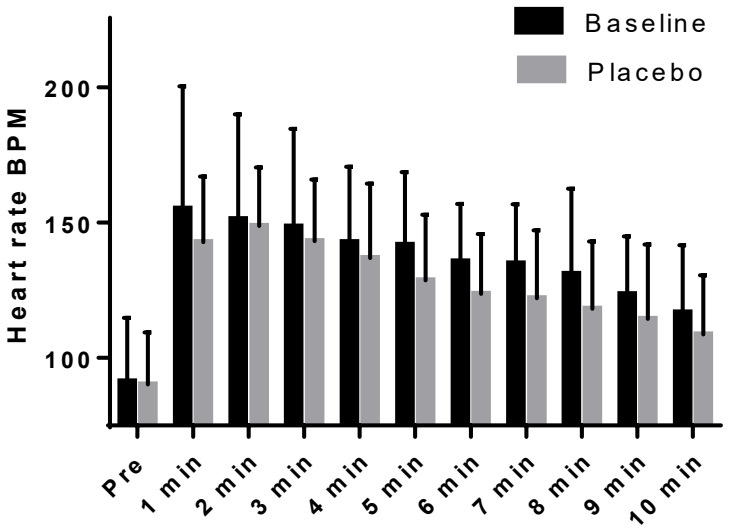
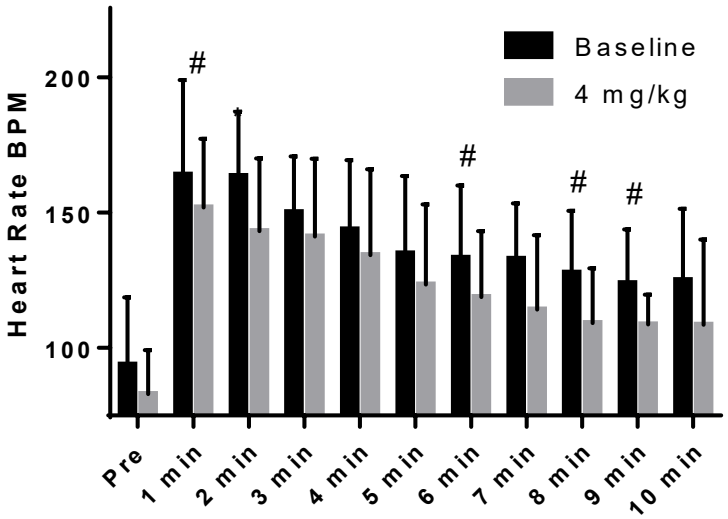
CanCog Car Ride Model

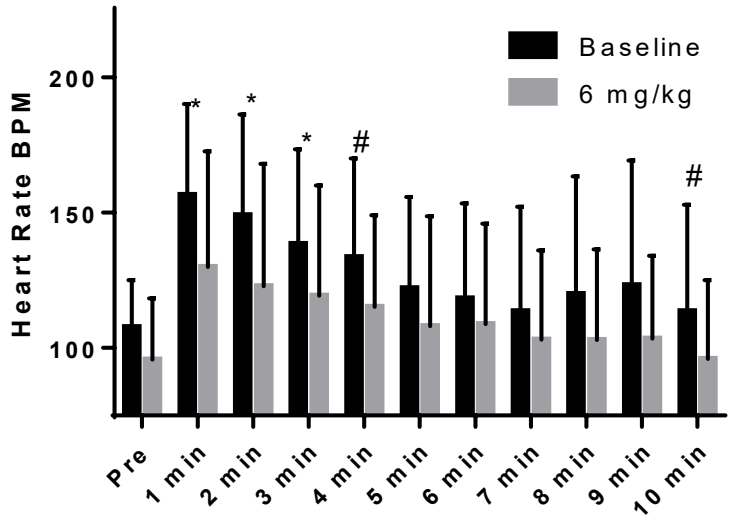
Heart Rate:



Placebo



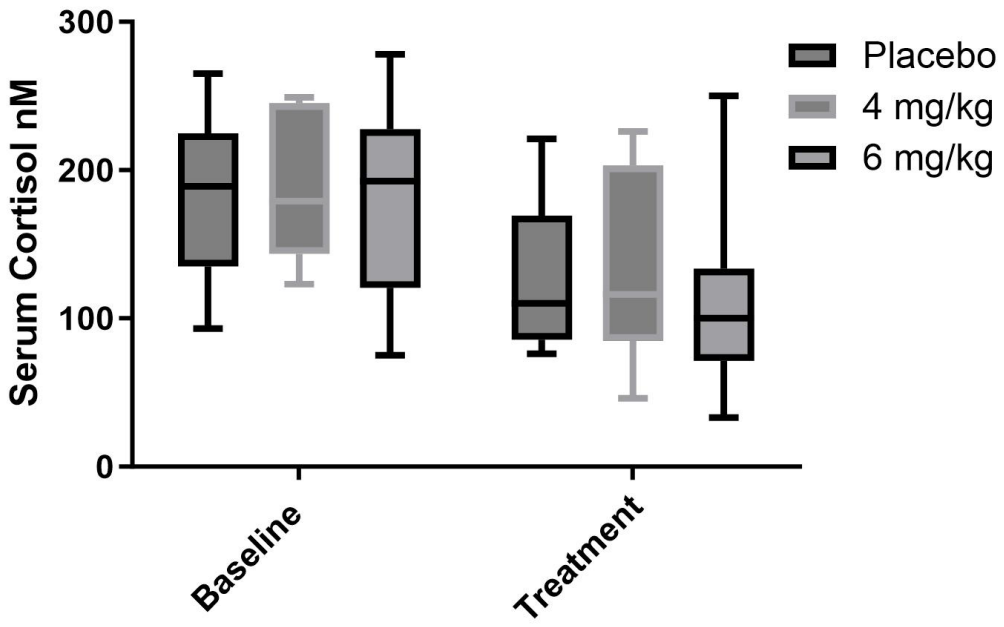
4mg/kg



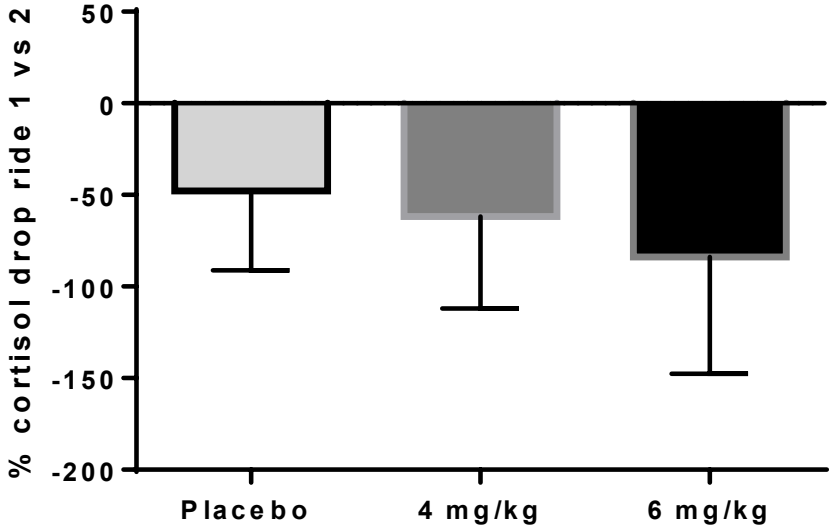
6mg/kg

CanCog Car Ride Model

Cortisol:



Serum Cortisol

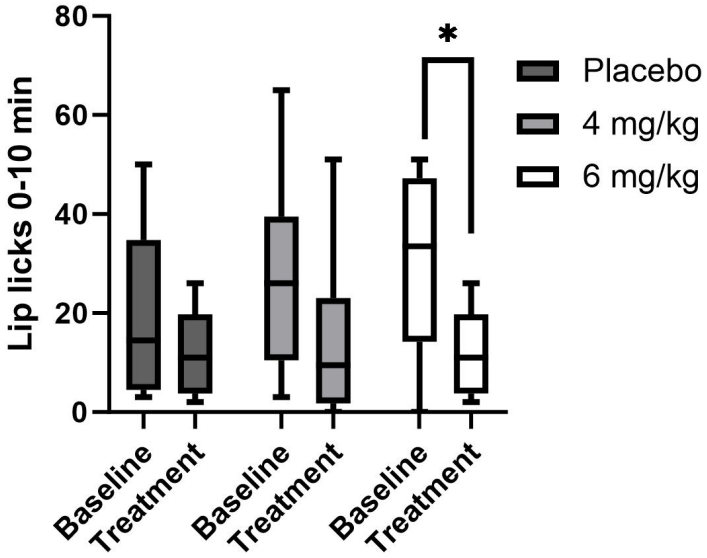


% Drop in Cortisol Ride 1 vs 2

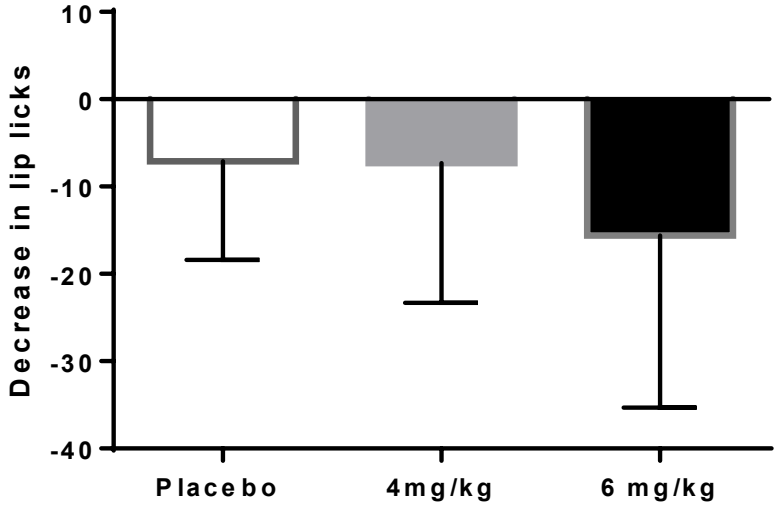
CanCog Car Ride Model

Lip Licking:

P = 0.012
Cohen's D:
4 mg/kg = 0.26 - weak
6 mg/kg – 0.70 - mod/strong



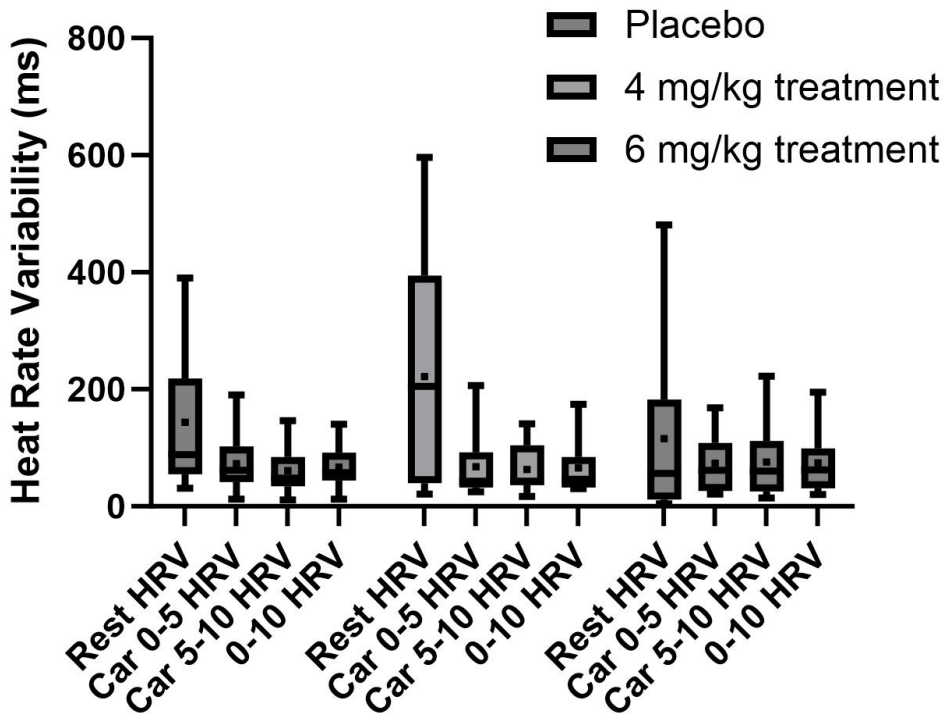
Number of Lip Licks in 10 mins



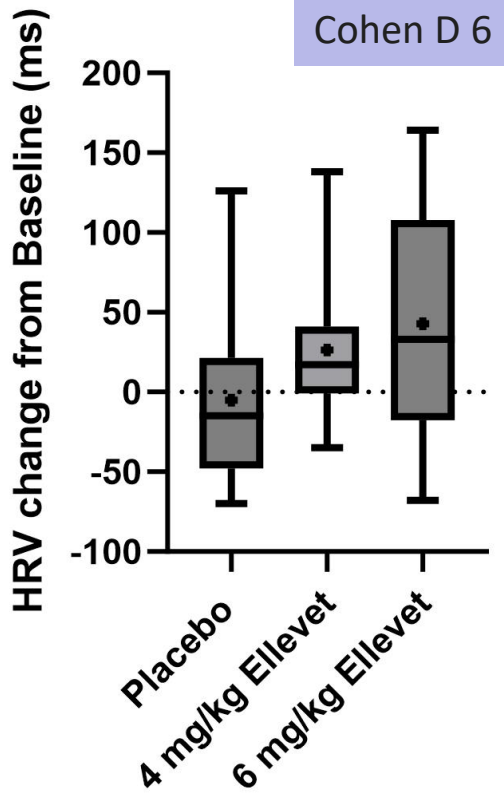
Decrease in Lip Licks

CanCog Car Ride Model

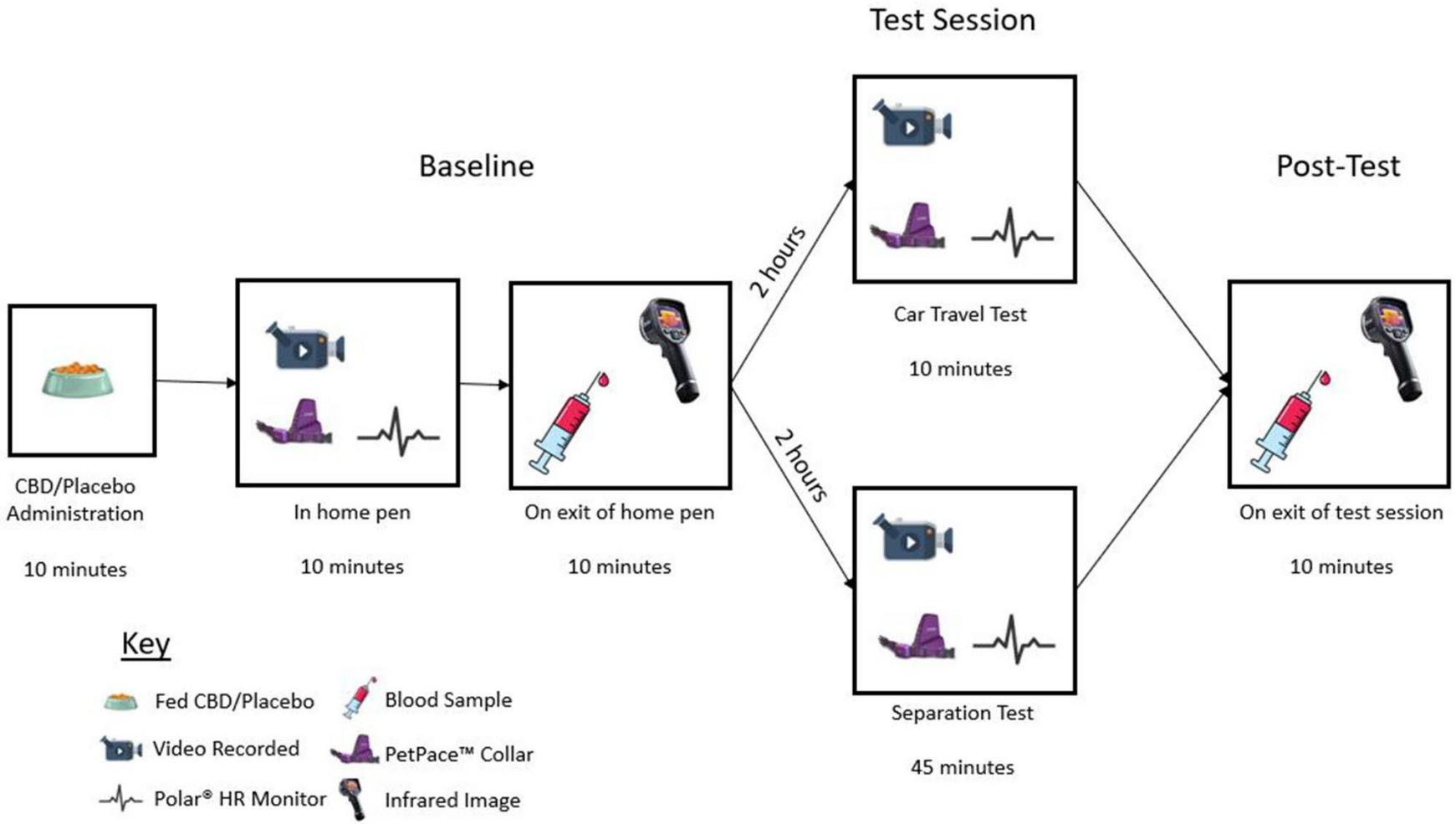
Heart Rate Variability:



P = 0.18
Cohen's D 4 mg/kg = 0.59 - mod
Cohen D 6 mg/kg = 0.71 - strong



A single dose of cannabidiol (CBD) positively influences measures of stress in dogs during separation and car travel



Alysia B. G. Hunt, Hannah E. Flint, Darren W. Logan and Tammie King*
Waltham Petcare Science Institute, Waltham on the Wolds, United Kingdom

Traditional Measures of Anxiety

Serum Measures

- Cortisol
- Serum glucose
- Serum IgA

TABLE 2 Ethogram used to measure dog behavior during the separation and car travel tests.

Behavior	Type		Definition
Repetitive pacing/circling	State	Start	Repeats behavioral sequence 2 or more times without a specific goal, following a fixed route. May pause for up to 2 s
		Finish	Dog ceases the repetitive nature of the movement or begins a different behavior
Panting	State	Start	Increased shallow respiration through an open mouth, may have tongue out (70)
		Finish	Mouth is closed—normal breathing resumes
Whining	State	Start	Dog produces sound such as whines, whimpers, and yelps originating from the throat and mouth
		Finish	Sounds production ceases
Barking	Event		Head and lips forward, mouth opening and shutting repeatedly to emit a large, sharp, short sound from the throat (84)*
Howling	Event		Raised muzzle perpendicular to ground and emits a long drawn out sound through semi-closed jaw (84)*
Play behavior	State	Start	Interaction (e.g., mouthing/pawing) with toys and/or box whilst exhibiting soft/relaxed body language
		Finish	Dog ceases behavior
Digging	State	Start	Mouth/front paws and claws used to attempt movement/displacement of substrate other than external door
		Finish	Dog ceases behavior
Escape behavior	State	Start	Tries to dig, bite, or scratch at the external door—not directed at themselves
		Finish	Dog ceases behavior
Elimination	Event		Squat/leg raised in order to urinate and/or hind end lowered and back arched in order to defecate (84)*
Vomiting	Event		Open mouth and retch causing vomit from the mouth
Yawning (car test only)	Event		An involuntary take of breath through a wide-open mouth (70)
Lip licking (car test only)	Event		Dog flicks tongue around the outside of mouth, on lips and/or quickly over the nose

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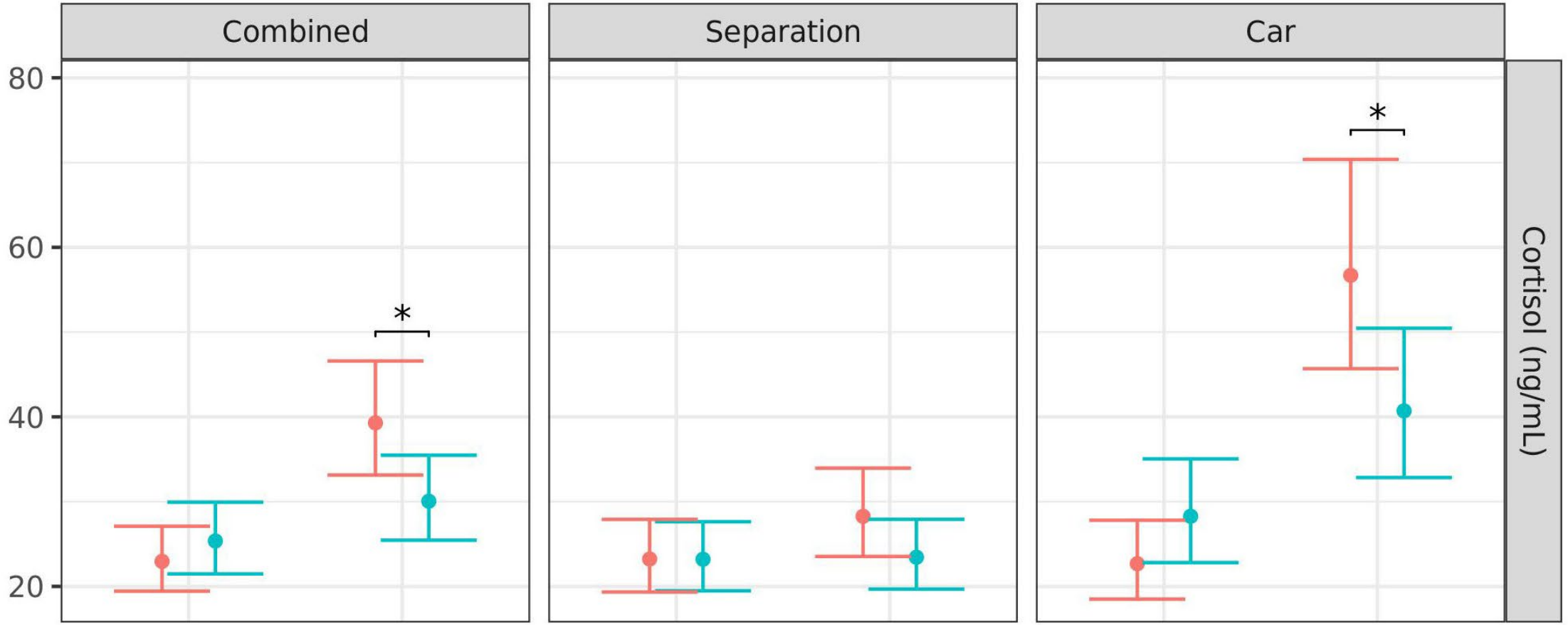
TABLE 1 Terms included in the Qualitative Behavioral Assessments (QBA) used to measure dog behavior during baseline, the separation, and car travel tests.

Term	Definition
Anxious	Worried, unable to settle or cope with the environment, apprehensive
Alert	Vigilant, inquisitive, on guard
Calm	Absent of strong positive/negative emotions
Comfortable	Without worries, settled in environment, peaceful with external stimuli
Depressed	Dull, sad demeanor, disengaged from and unresponsive to the environment, quiet, apathetic
Explorative	Confident in exploring the environment or new stimuli, investigative
Fearful	Timid, scared, shows postures typical of fear
Lethargic	Sluggish, inactive, unresponsive or slow to respond to external stimuli
Nauseous (car test only)	Salivating, lip licking, facial tension, excessive swallowing, retching, hunched body posture
Nervous	Uneasy, agitated, shows fast arousal, unsettled, restless, hyperactive
Reactive	Responsive to external stimuli
Relaxed	Easy going, calm with no visual evidence of tension in the body
Restless	Unable to rest or relax
Sad	Unhappy, downcast
Stressed	Tense, shows signs of distress
Tense	Stiff, rigid posture, on edge
Uncomfortable	Uneasy, nervous, tense, restless

control and nine treatment) experiencing car travel. Dogs in the separation group were habituated to the testing room until they were deemed comfortable in the environment with their experienced handler present. Dogs in the car travel group were trained to enter a crate within the car *via* a ramp or box setup voluntarily and habituated to the crate.

- Three raters with good interrater reliability for videos
  - 10 minutes for car ride
  - First, Middle, last 5 minutes of 45 minutes separation
- Assessed QBA
  - Differences in sadness, stressed, tense and uncomfortable and more explorative in separation only.

Serum Cortisols



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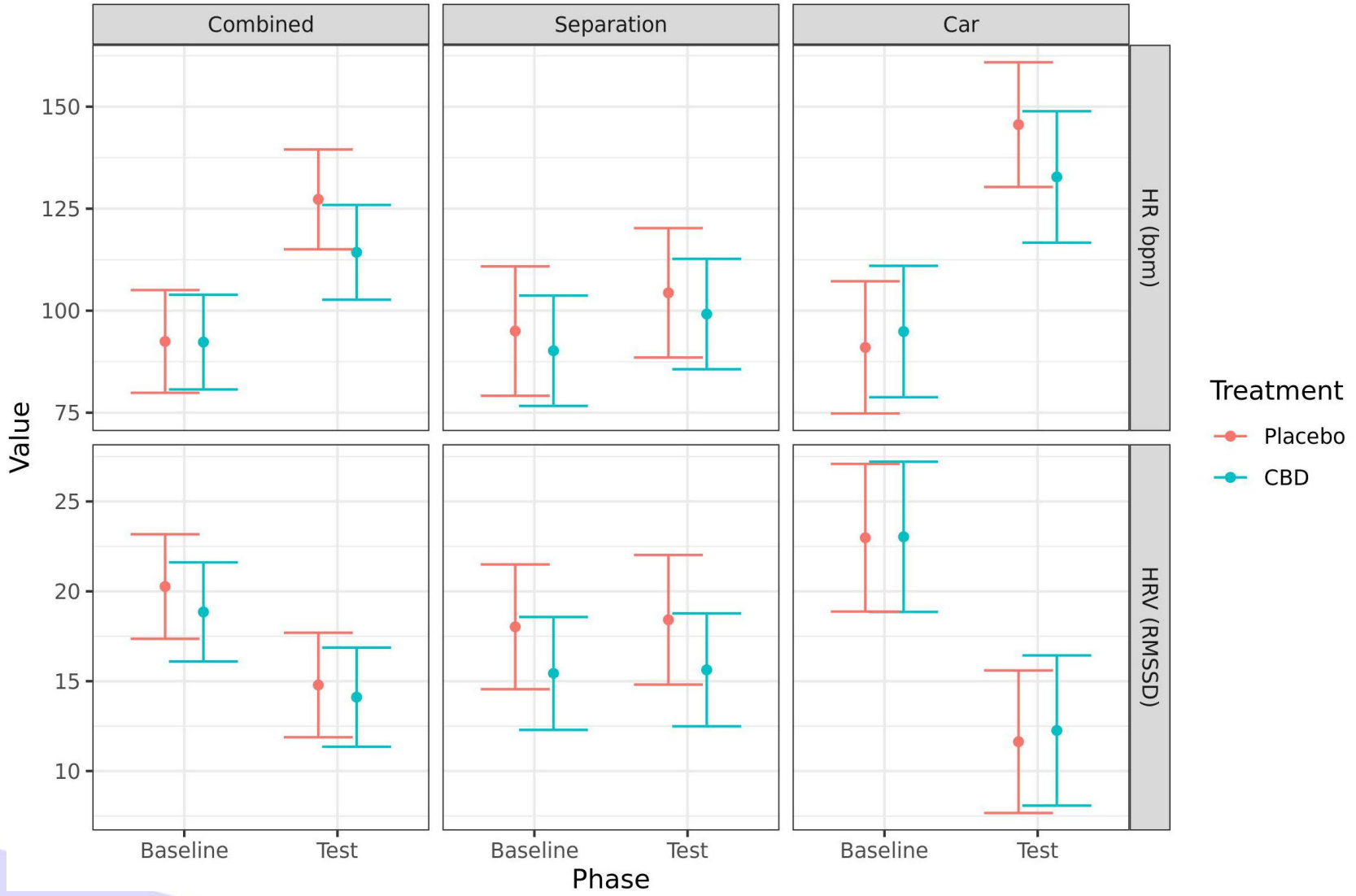
Heart Rate and Variability



Typical Measures of Anxiety

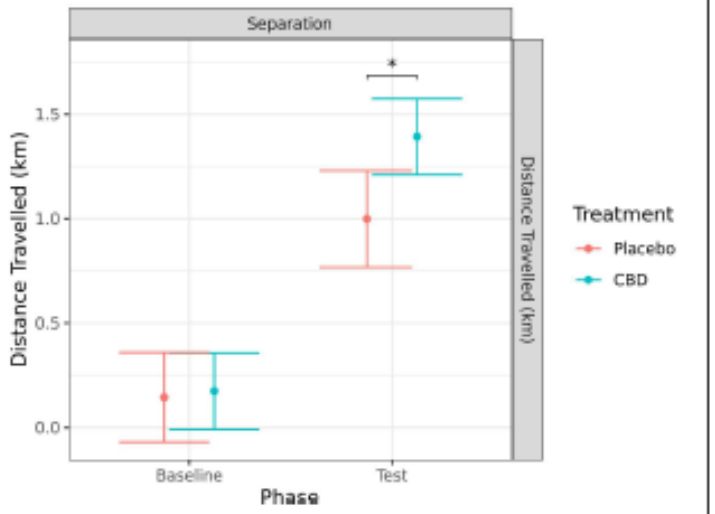


FIGURE 10
Predicted mean (95% CI) distance traveled (km) generated by the Polar® device for dogs given CBD or placebo at each phase of testing (baseline and test) during the separation test. Asterisks indicate significant differences between treatment groups within each phase. *$p < 0.05$.

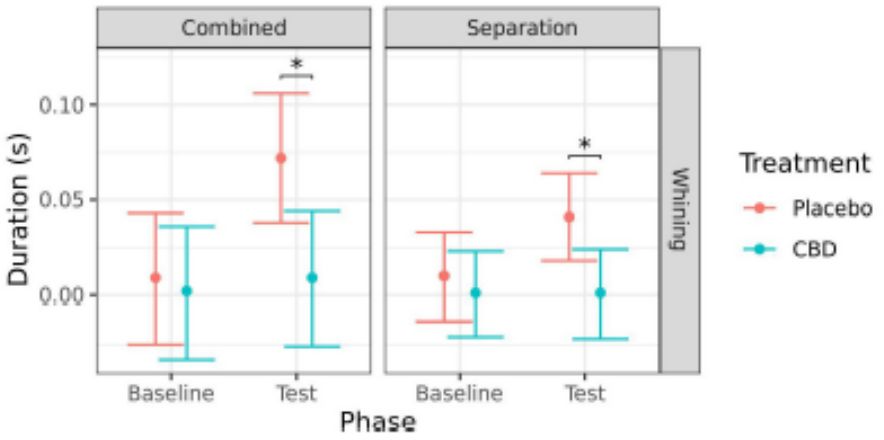


FIGURE 7
Predicted mean (95% CI) duration of time spent whining(s) for dogs given CBD or placebo at each phase of testing (baseline and test) based on models analyzing both stress tests combined or separation test. Asterisks indicate significant differences between treatment groups within each phase. *$p < 0.05$.

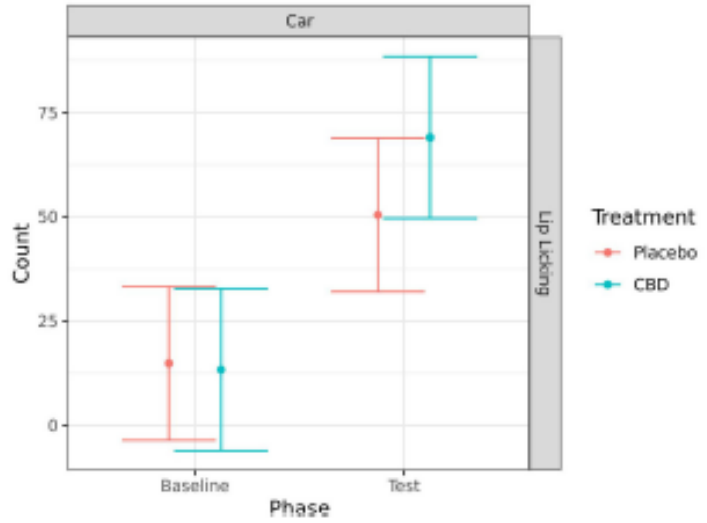


FIGURE 8
Predicted mean (95% CI) number of lip licks for dogs given CBD or placebo at each phase of testing (baseline and test) during the car test. No significant differences ($p < 0.05$) were identified between treatment groups within each phase.

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What needs to be done!

in the dogs tested (38, 83). Only three dog breeds were represented, all clinically healthy and living in a homogeneous environment, and they were not “owned” pets selected for displaying separation-related anxiety or travel-related stress. Therefore, confirming these research findings in pet dogs in traditional home environments will be beneficial. Doses >4 mg/kg, have caused mild side effects in tolerance studies (39), but testing the efficacy of lower or multiple dosing of CBD in the same stress paradigms may also be worthwhile.

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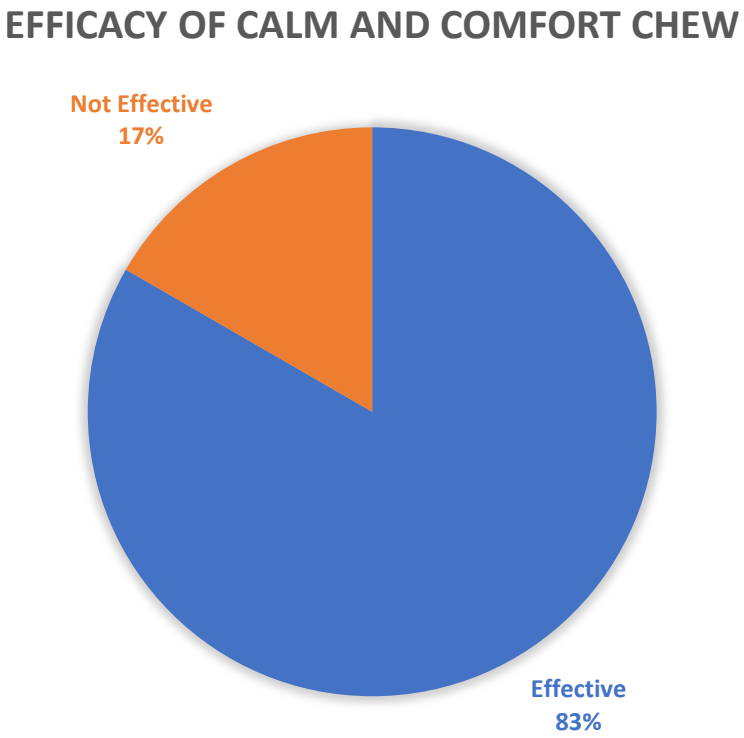
Why Do We Think it works?

- Real life study of 24 dogs with
  - Car ride anxiety
  - New person aversion
  - Noise phobia
- Observed dog under stimuli
- Provided around 3.5 mg/kg per dog.
- Looked for onset of effect
  - 4/24 within 30 minutes
  - 10/24 within 60 minutes
  - 6/24 within 120 minutes
- Still the leading indication based on sales.



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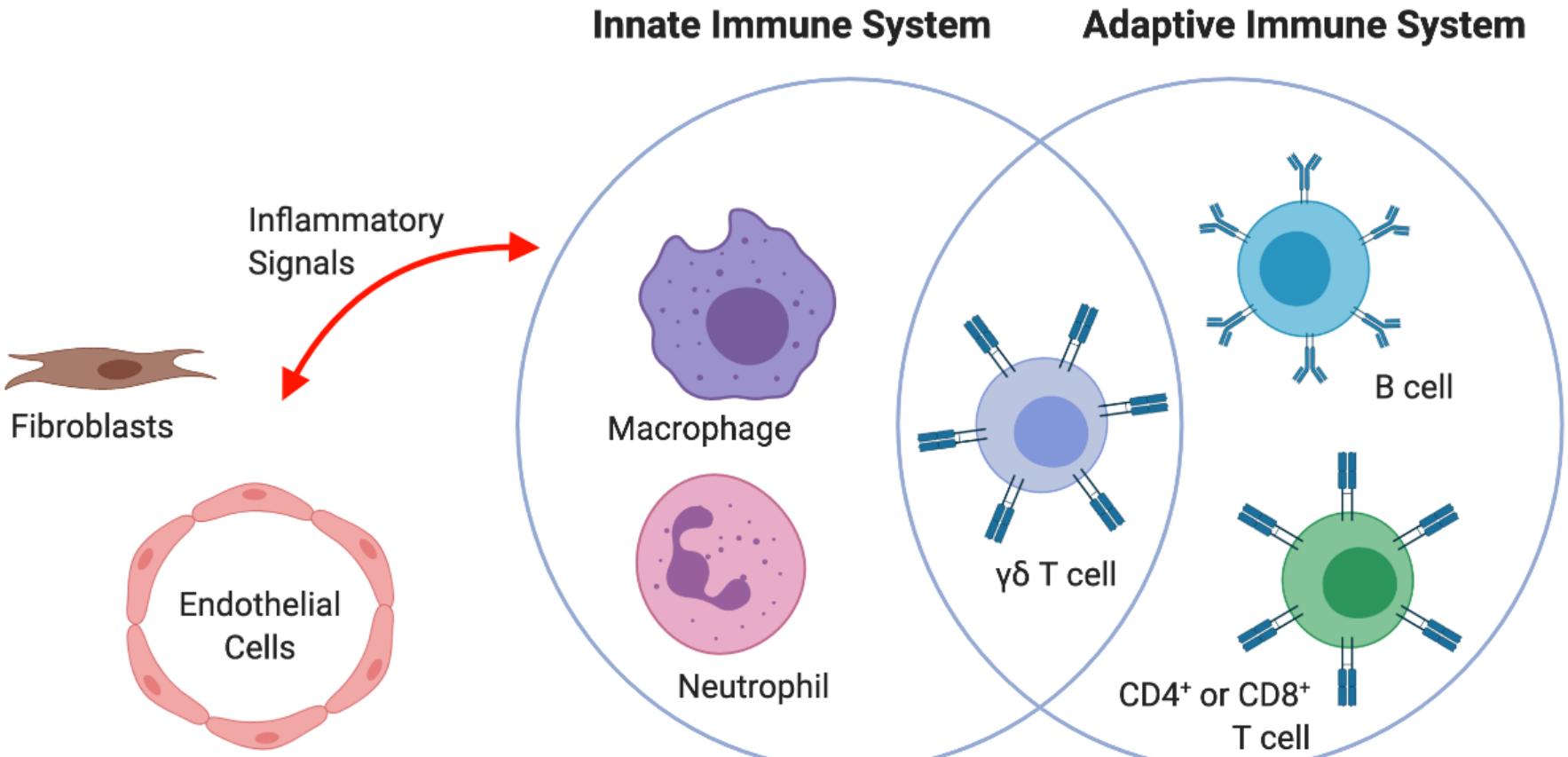
CBD and the immune system?



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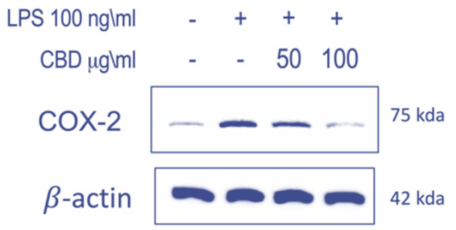
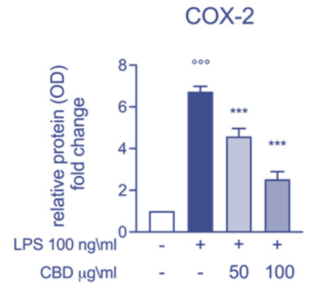
Immune Cell Work in Canine!

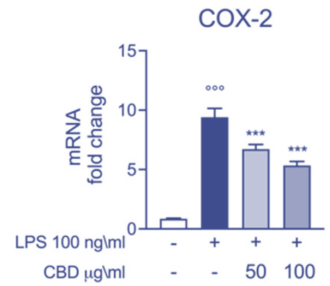
Article

Effect of Cannabidiol (CBD) on Canine Inflammatory Response: An Ex Vivo Study on LPS Stimulated Whole Blood

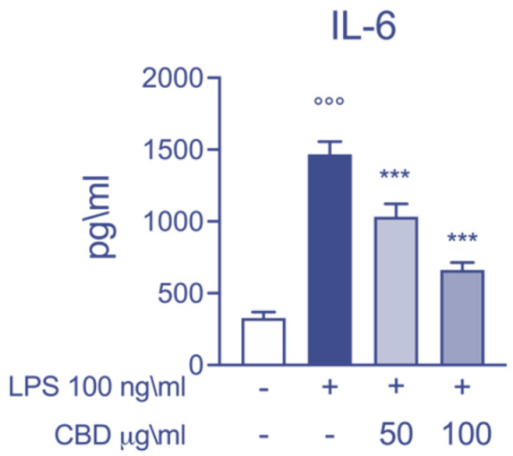
Enrico Gugliandolo [1], Patrizia Licata [1], Alessio Filippo Peritore [2], Rosalba Siracusa [2], Ramona D'Amico [2], Marika Cordaro [3], Roberta Fusco [2], Daniela Impellizzeri [2], Rosanna Di Paola [2,*], Salvatore Cuzzocrea [2,4,*], Rosalia Crupi [1,†] and Claudia Dina Interlandi [1,†]

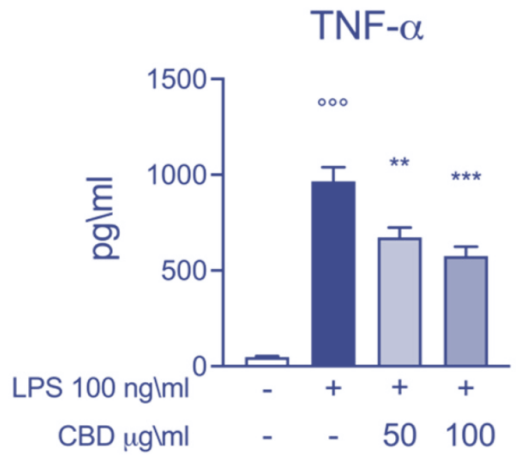






- Whole blood from 6 German Shepard Dogs
- Incubated with Vehicle control or CBD at 50 ug/mL of 100 ug/mL for 24 hrs
- Then stimulated with LPS to induce cytokine production
- Assessed IL-6, TNF alpha, Nf-KB and COX-2 expression
- This is 100 fold higher than we can reach in the plasma!





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Cannabinoid receptor type 1 and 2 expression in the skin of healthy dogs and dogs with atopic dermatitis

Luca Campora, DVM, PhD; Vincenzo Miragliotta, DVM, PhD; Emanuele Ricci, DVM, PhD; Luigia Cristino, Biol D, PhD; Vincenzo Di Marzo, Chem D, PhD; Francesco Albanese, DVM; Maria Federica della Valle, MSc; Francesca Abramo, DVM

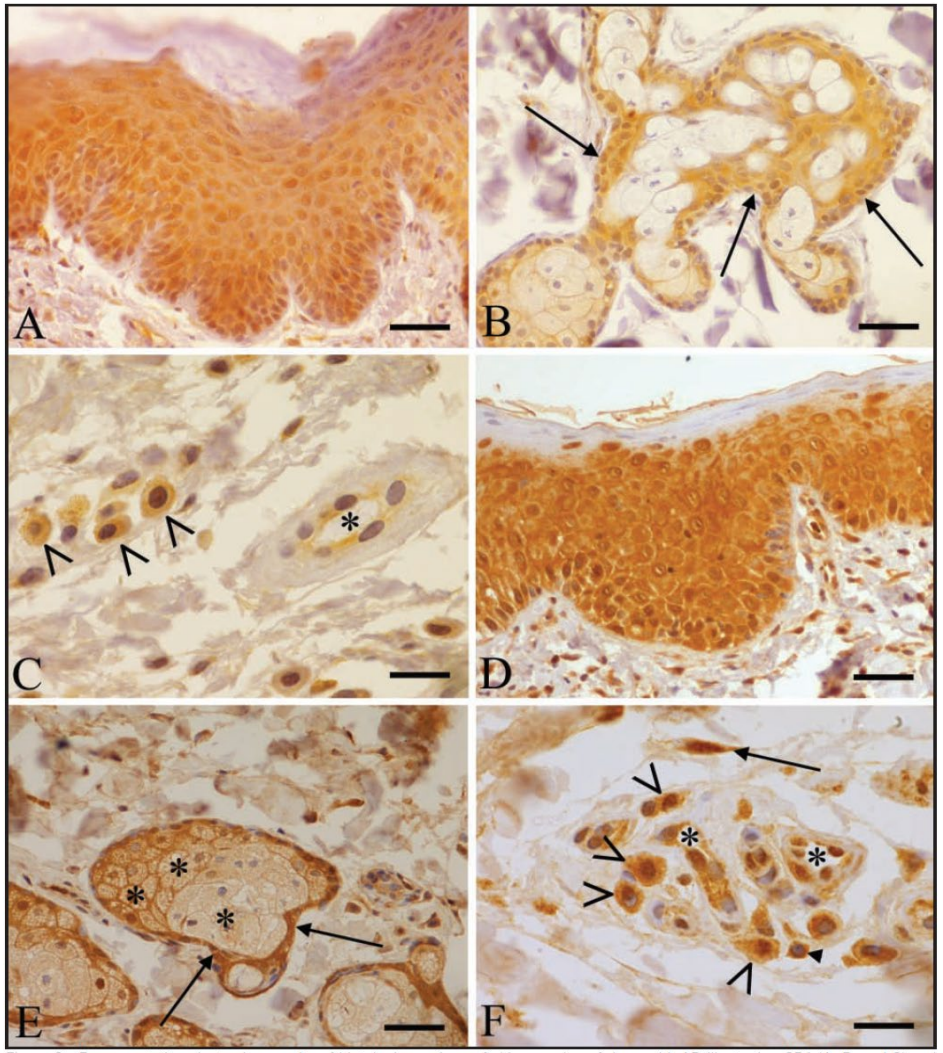

Figure 3—Representative photomicrographs of histologic sections of skin samples of dogs with AD illustrating CB1 (A, B, and C) and CB2 (D, E, and F) immunoreactivity. A—Notice CB1 immunoreactivity in suprabasal and granular layers of epidermis. Bar = 50 µm. B—Notice CB1 immunoreactivity in reserve cells of hypertrophic sebaceous glands (arrows). Bar = 50 µm. C—Notice CB1 immunoreactivity in perivascular cells with mast cell morphology (open arrowheads) and endothelial cells lining the lumen of a small blood vessel (asterisk). Bar = 125 µm. D—Notice CB2 immunoreactivity in basal, suprabasal, and granular layers of epidermis. Bar = 50 µm. E—Notice CB2 immunoreactivity in peripheral sebaceous reserve cells (arrows) and centrally located mature sebocytes (asterisks). Bar = 50 µm. F—Notice CB2 immunoreactivity in perivascular cells with mast cell morphology (open arrowheads), endothelial cells lining lumina of small blood vessels (asterisks), a fibroblast-like dermal cell (arrow), and a lymphocyte (small solid arrowhead). Bar = 125 µm.

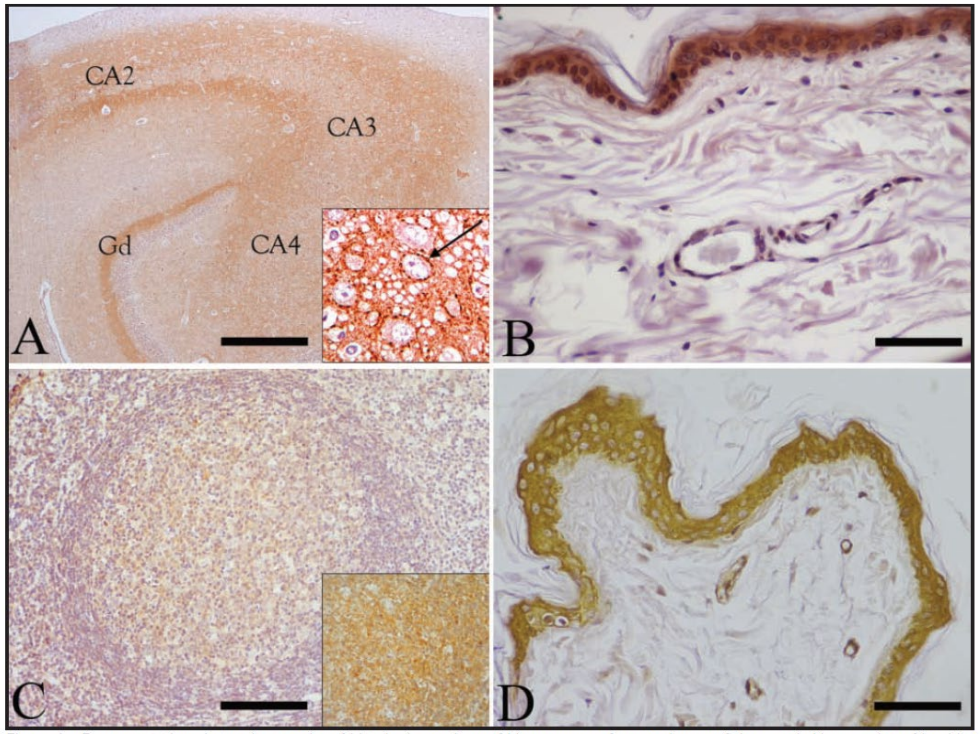

Figure 1—Representative photomicrographs of histologic sections of hippocampus from cadavers of dogs and skin samples of healthy dogs illustrating immunohistochemical staining for CB1 (A and B) and CB2 (C and D). A—Photomicrograph of a histologic section of hippocampus. Notice strong CB1 immunoreactivity in various regions of the hippocampus. Bar = 500 µm. Inset—Neuronal cell bodies in the neuropil of the pyramidal layer are surrounded by dot-like structures with CB1 immunoreactivity (arrow). B—Photomicrograph of a histologic section of skin. Notice CB1 immunoreactivity in keratinocytes in the basal and suprabasal epidermal layers. Bar = 50 µm. C—Photomicrograph of a histologic section of skin. Notice CB2 immunoreactivity in lymphocytes in the follicular B-cell regions. Bar = 100 µm. Inset—Lymphocytes in the follicular germinal center have CB2 immunoreactivity. D— Photomicrograph of a histologic section of skin. Notice CB2 immunoreactivity in basal and suprabasal epidermal keratinocytes. Bar = 50 µm. Gd = Dentate gyrus.

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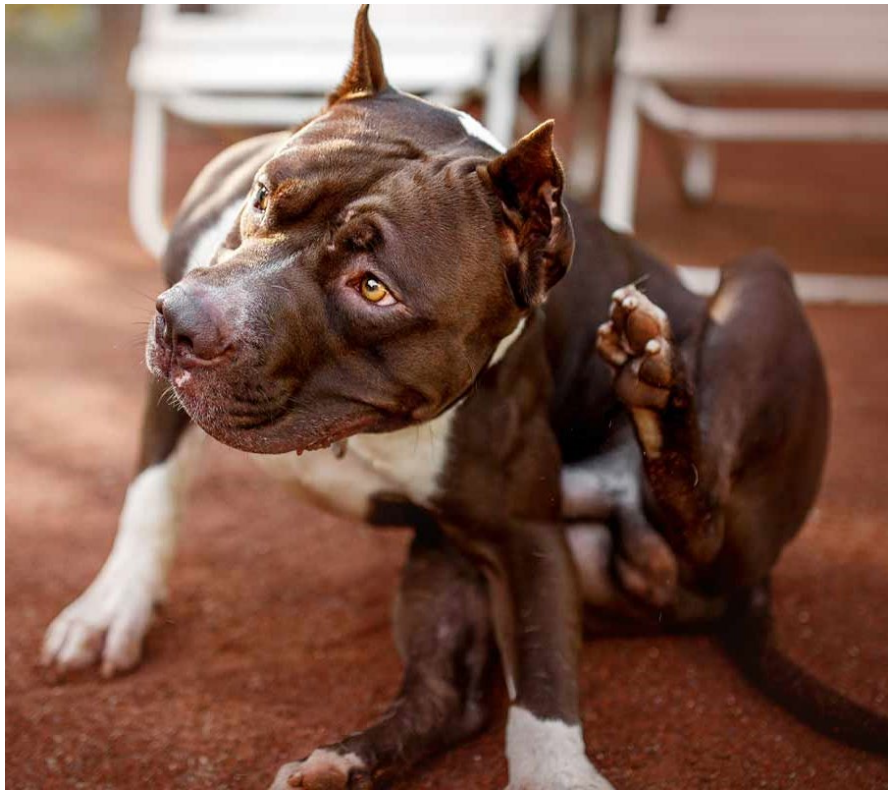
Receptors and Atopic Dermatitis?

- We currently do not understand the receptor biology as it relates to Cannabinoids in the skin
  - Immune Cells – Mast Cells, lymphocytes, plasma cells, APC (monocytes)
  - Keratinocytes and the adnexal structures.
  - CB1 and CB2 receptors upregulated in skin of atopic dogs
  - Receptors CB2, GPR55, TRPV1, TRPM, 5HT, all CBD, CBDA, CBG, CBGA targets
  - Dr. Roberto Chiochetti - IHC
- Do cannabinoids alter inflammation and/or can they alter the pruritus?



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Atopic Dermatitis Study!

- Accepted and published in Vet Derm!
- Double-blinded placebo controlled
  - 17 Treatment Group/ 12 Placebo
  - 2 mg/kg BID for one month
  - Assessing CADESI-04 scoring (vet)
  - VAS pruritus (owner)
  - Owner satisfaction
  - Serum Cytokines
  - IL-34 - associated with CADESI score
  - IL-31 – associated with pruritus
  - CBC and serum chemistry
  - Serum cannabinoids

ADAS
ANIMAL DERMATOLOGY
& ALLERGY SPECIALISTS

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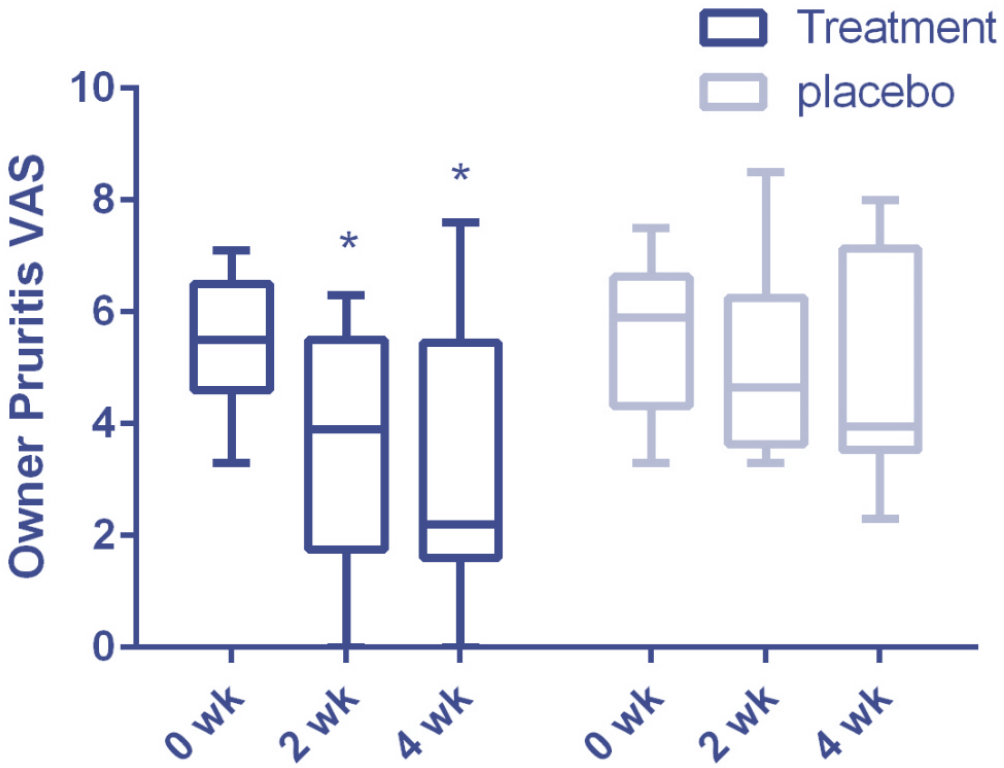
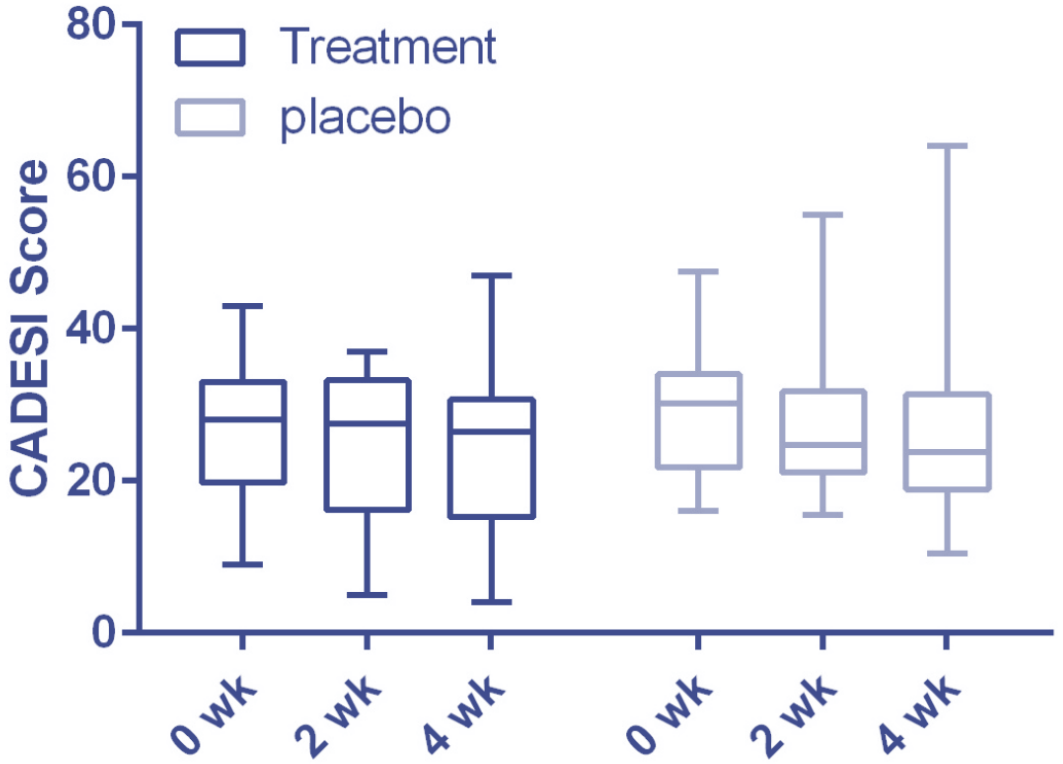
VAS and CADESI scores





Fisher's Exact = 0.0256	Responded/ would continue	No Response
Treatment	10	7
Placebo	2	10

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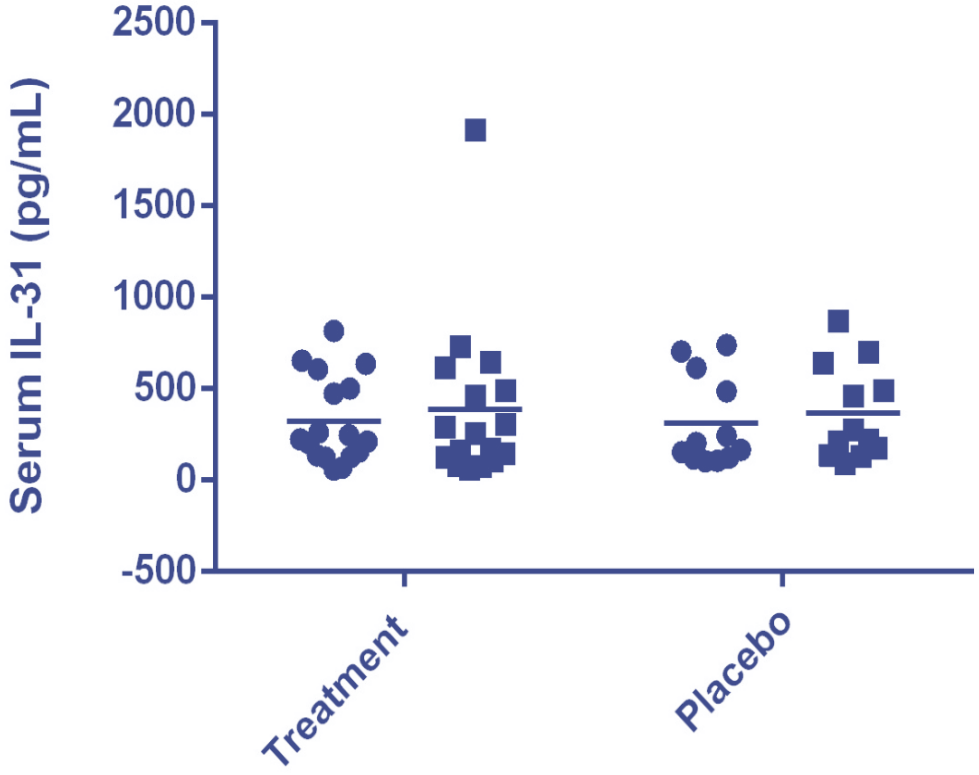
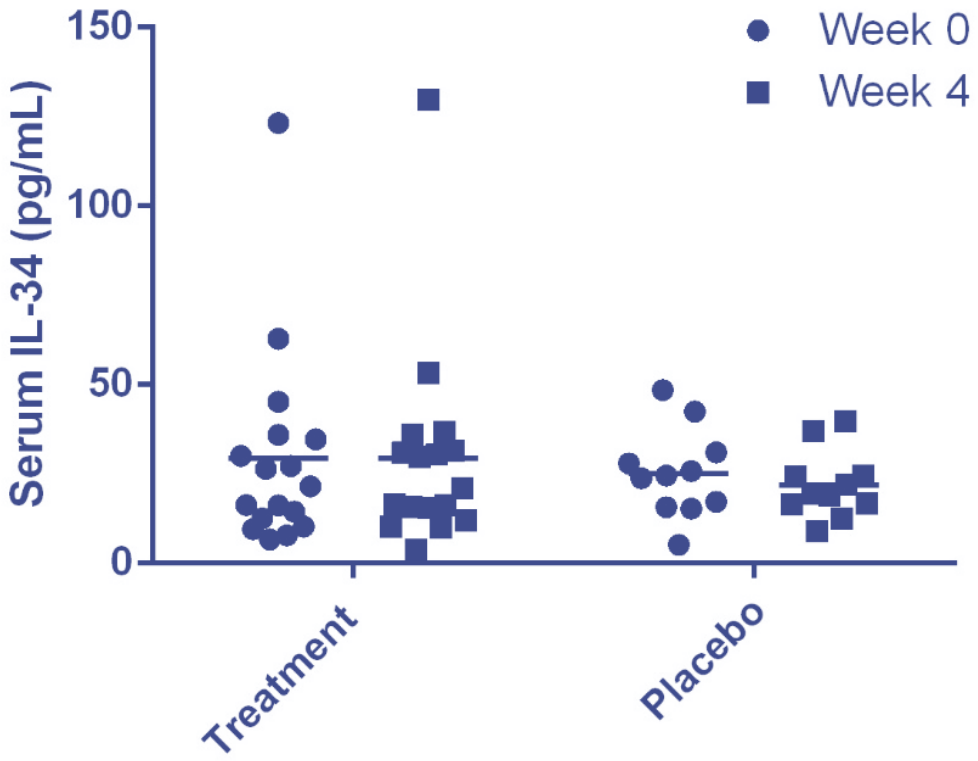
IL-34 (CADESI) and IL-31 (Pruritus)





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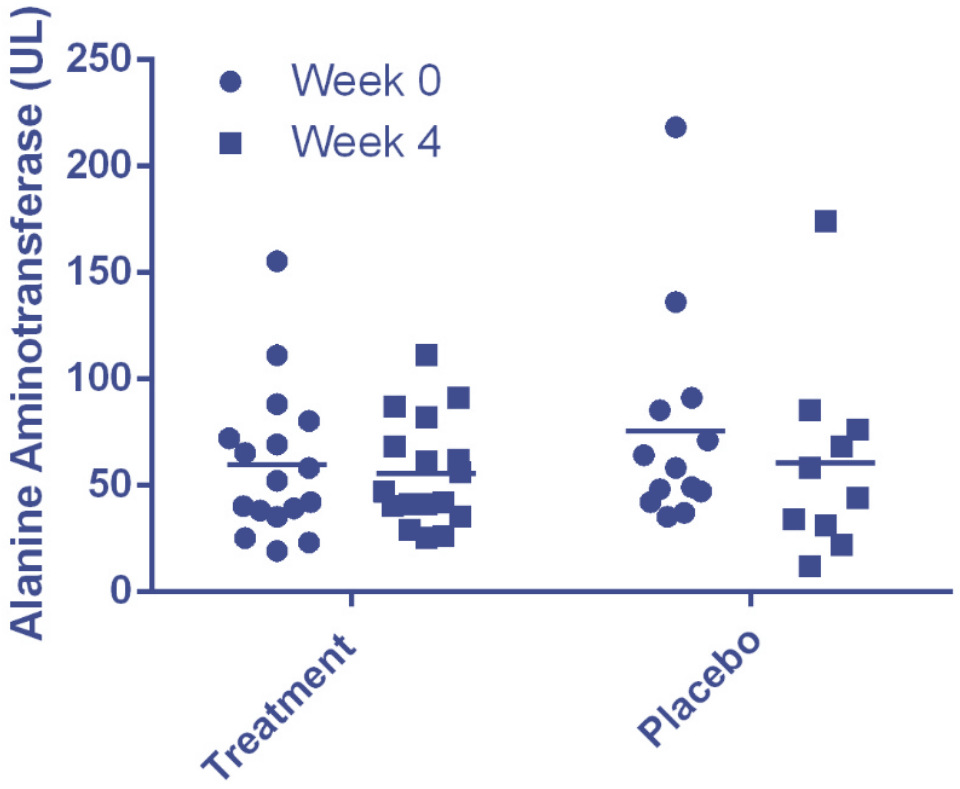
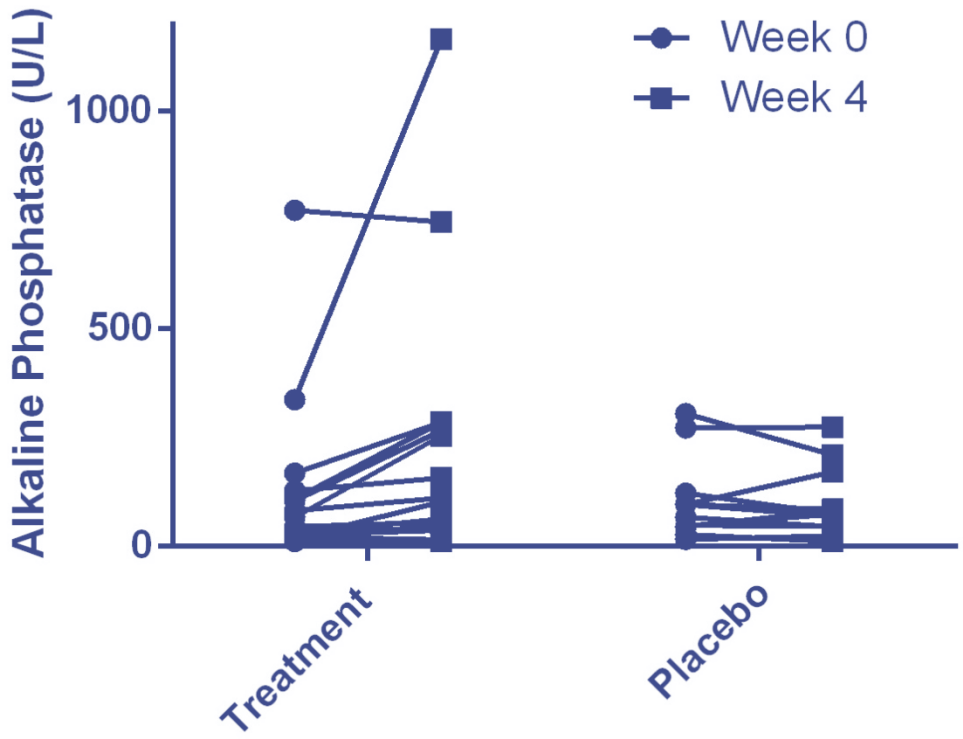
Liver Enzymes – ALP and ALT





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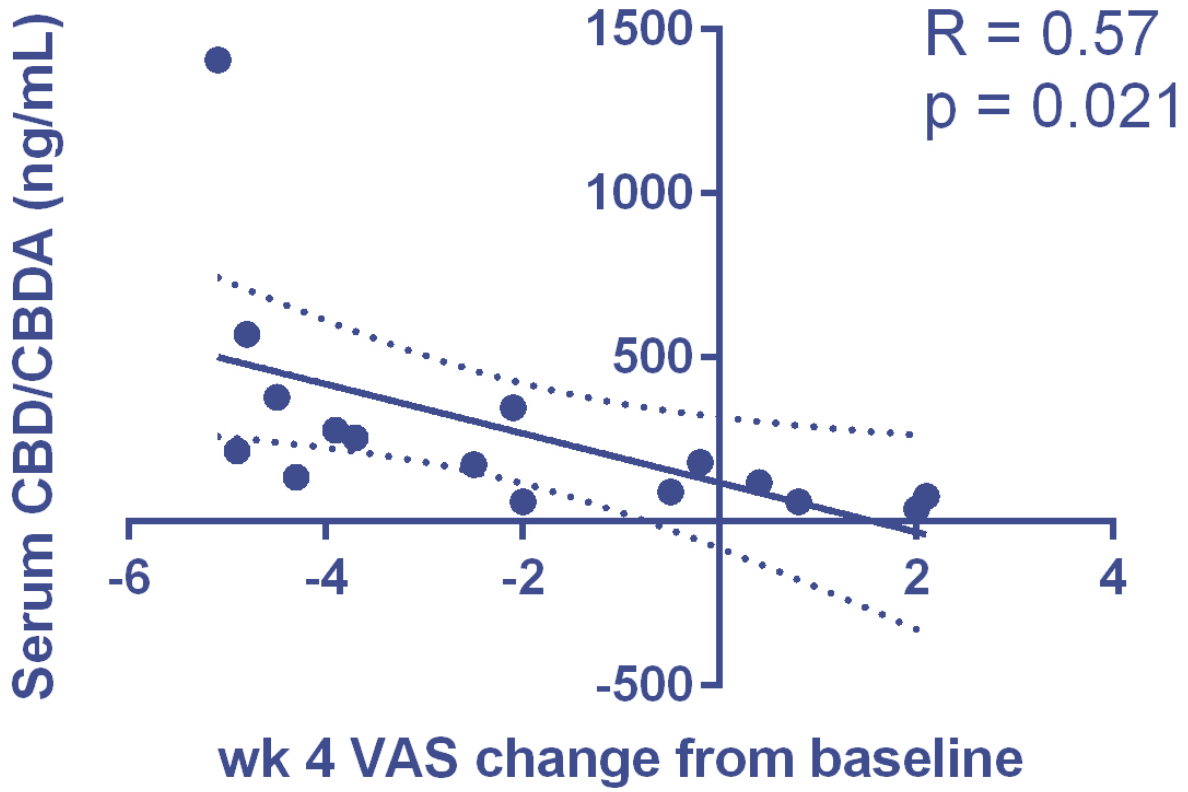
Regression of CBD/CBDA and VAS change:



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CBD and Pruritus?

Feeding Cannabidiol (CBD)-Containing Treats Did Not Affect Canine Daily Voluntary Activity

Elizabeth M. Morris[1], Susanna E. Kitts-Morgan[2], Dawn M. Spangler[2], Jessica Gebert[2], Eric S. Vanzant[1], Kyle R. McLeod[1] and David L. Harmon[1]*

[1] Department of Animal and Food Sciences, University of Kentucky, Lexington, KY, United States, [2] College of Veterinary Medicine, Lincoln Memorial University, Harrogate, TN, United States

…….During the PM session, dogs receiving 4 mg CBD/kg BW/d tended (P=0.091) to be less active than control. During AM and PM sessions, CBD reduced scratching compared with CON (P=0.030). CBD did not affect activity duration during exercise periods (P=0.143). These results indicate that when supplemented with up to 4 mg CBD/kg BW/d, CBD does not impact the daily activity of adult dogs but may exert an antipruritic effect.

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Summary

- We are understanding how CBD/CBDA can be used to mitigate refractory seizures -40% response rate at 2 mg/kg and up
  - Can be used with other medication safely in epilepsy
  - Watch for somnolence primarily
- Anxiety and Behavioral issues are possible venues
  - Hemp CBD/CBDA products may be used in situational events
  - Dose may be higher 3-6 mg/kg
  - No adverse events are noted at higher doses.
  - Still leading reason for use in clients – further real-life studies are needed!
- Don’t forget the atopic itchy dogs!

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Upcoming and ongoing research!

Selection of areas of current investigation by ElleVet Sciences

- Anxiety –separation anxiety
- Oncology – QoL during Chemo
- Immune cell regulation
- Feline OA
- Feline interstitial cystitis
- Horse OA
- Zoo and exotics OA

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Red Bank Veterinary Hospitals
Summit Ridge Farms
Florida Veterinary Behavior Service
LMU Ludwig-Maximilians-Universität München
Universiteit Utrecht
University of Miami Miller School of Medicine
The University of Tennessee College of Veterinary Medicine
Johns Hopkins Medicine

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Questions or comments?

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