Evaluation of the Effect of Cannabidiol on Naturally Occurring Osteoarthritis-Associated Pain: A Pilot Study in Dogs

Sebastian Mejia, DVM*, Felix Michael Duerr, DVM, MS, DACVS-SA, DECVS, DACVSMR, Gregg Griffenhagen, DVM, MS, DACVAA, Stephanie McGrath, DVM, MS, DACVIM (Neurology)

ABSTRACT

The objective of this study was to provide preliminary data describing the safety and effect of cannabidiol (CBD) for symptom relief of canine osteoarthritis-associated pain in a clinical setting using objective outcome measures. Twenty-three client-owned dogs with naturally occurring osteoarthritis of appendicular joints completed this prospective, double-blinded, crossover, placebo-controlled study. Baseline data were acquired for 4 wk, followed by random allocation to either placebo or CBD treatment for 6 wk, followed by 6 wk with the opposite treatment. Outcome measures included objective gait analysis, activity counts (via accelerometry) and clinical metrology instruments. There were no differences noted between groups at any time point for any of the recorded outcome measures. Adverse events associated with CBD administration included elevation in liver enzymes (n = 14) and vomiting (n = 2). (*J Am Anim Hosp Assoc* 2021; 57:■■■–■■■. DOI 10.5326/JAAHA-MS-7119)

Introduction

Osteoarthritis (OA) is the most common joint disorder in both human and veterinary medicine, leading to subsequent impaired mobility and functional disability.[1,2] Its incidence in veterinary medicine has been reported between 2.5 and 20% of the canine population over 1 yr of age being affected.[1,3] A recent large population study reported musculoskeletal disease and inability to stand as the leading causes of death (overcoming neoplastic disease) in German shepherd dogs.[4] Given the degenerative and progressive nature of the condition, treatment options are limited, and a multimodal approach for the management of clinical signs associated with OA is frequently pursued. Treatment strategies include a combination of surgical and nonsurgical interventions.[5] Nonsteroidal anti-inflammatory drugs (NSAIDs) are a frequently used

From the Department of Clinical Sciences, James L. Voss Veterinary Teaching Hospital, Colorado State University, Fort Collins, Colorado.

Correspondence: felix.duerr@colostate.edu (F.M.D.)

AC (activity count); %BWD (percentage of body weight distribution); CB1 (cannabinoid receptor type 1); CB2 (cannabinoid receptor type 2); CBD (cannabidiol); CBD-PL (CBD treatment followed by placebo treatment group); CBPI (Canine Brief Pain Inventory); CMI (clinical metrology instrument); NSAID (nonsteroidal anti-inflammatory drug); OA (osteoarthritis); PIS (pain interference score); PL-CBD (placebo treatment followed by CBD treatment group); PSW (pressure-sensitive walkway); PVF% (peak vertical force normalized by body weight); THC (tetrahydrocannabinol)

*S. Mejia's present affiliation is the Surgery Department, LeadER Animal Specialty Hospital, Cooper City, Florida.

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pharmacologic treatment option for pain control and are considered standard of care. Although recent systematic reviews suggest that severe clinical adverse events are rare,[6] there is concern regarding the long-term use of NSAIDs among veterinarians.[7] Unfortunately, other medications appear to be less effective in canine patients,[5] leading to a need for alternative medications that are safe, effective, and easy to administer.

Recently, there has been an increase in the use of cannabinoids for multiple conditions, including epilepsy and OA in both veterinary and human medicine.[8,9] Cannabinoids are classified into three major categories based on their origin: endocannabinoids, phytocannabinoids, and synthetic cannabinoids. Phytocannabinoids, like tetrahydrocannabinol (THC) and cannabidiol (CBD), are generally harvested from the female plants of *Cannabis sativa*.[8] Cannabidiol acts via the endocannabinoid system, a biochemical signaling system composed of receptors, ligands, and signaling enzymes.[10] Two of the most investigated receptors are cannabinoid receptor type 1 (CB1) and cannabinoid receptor type 2 (CB2). CB1 receptors are mostly located in the central nervous system and have effects on memory, appetite, and neuronal excitability, whereas CB2 receptors are located largely on immune cells (i.e., macrophages, mast and glial cells) where they act to inhibit production of proinflammatory cytokines.[11] The pain modulatory effect of CBD is complex, but altering the activity of CB1 and CB2 receptors or the fatty acid amide hydrolase enzymes likely plays a major role.[8,11]

Although cannabinoids have been used for the treatment of symptoms associated with chronic pain and other conditions in humans for centuries, scientific evidence is still limited.[8] Recently, the DEA rescheduled a CBD-based product (Epidiolex) approved by the FDA as a Schedule V substance. Additionally, the 2018 federal Farm Bill descheduled industrial hemp (defined as plants with $<0.3\%$ THC content by dry weight).[12] Experimental studies using rat and mouse models evaluating CBD for the amelioration of clinical symptoms associated with OA have shown promising results.[13,14] A canine study confirmed the presence of two known endocannabinoids (N-Arachidonoylethanolamide and 2-Arachynonyl Glycerol) within the synovial fluid of arthritic stifles,[15] suggesting endocannabinoid system activity associated with this condition in dogs. Recent clinical studies have investigated the safety of CBD for dogs and cats[16–19] and found clinical improvement in clinical metrology instruments (CMIs) as well as a decrease in the veterinary assessment of pain and analgesic requirements.[16,18–20] However, to date, there is a lack of sufficient clinical research evaluating the effect of CBD on OA-associated pain using objective outcome measures, such as ground reaction forces and accelerometry.

The objective of this study was to provide preliminary data describing the short-term safety and effect of CBD on symptom relief of canine OA-associated pain in a clinical setting using objective outcome measures.

Materials and Methods

This was a prospective, randomized, crossover, double-blinded, placebo-controlled clinical trial. The protocol was approved by the Clinical Review Board of the Colorado State University Teaching Hospital (VCS #2016-073), and owner consent was obtained before enrollment. Client-owned dogs with naturally occurring OA of the carpus, elbow, shoulder, tarsus, stifle, or hip joint of any breed or sex presenting to the Colorado State University Veterinary Teaching Hospital were eligible for participation. Inclusion criteria were a body weight >15 kg; >3 yr of age; unremarkable general physical examination, complete blood count, and serum chemistry; a subjectively identifiable lameness (≥ 2 and <5 on a 0–5 scale; supplementary Table I) as determined by a board-certified veterinary surgeon; radiographically confirmed OA (within 6 mo of enrollment) consistent with the observed lameness; and a Canine Brief Pain Inventory (CBPI) pain severity score and pain interference score (PIS) ≥ 2 for each.[21] Exclusion criteria included palpable instability of the shoulder or stifle joint (dogs with chronic, stable cranial cruciate ligament disease were eligible), clinically significant orthopedic disease (other than OA), neoplasia, evidence of neurologic disease, any previous orthopedic surgical procedure or any intra-articular injection performed within 6 mo before enrollment, and concurrent treatment with any cannabis product at the time of evaluation.

At the time of enrollment, treatment strategies for each patient were discussed and changes to the current treatment regimen were suggested. Owners were given the option to continue enrollment if they elected to not pursue any of the suggested changes or to postpone enrollment until these changes were instituted. All patients were required to be on a consistent treatment regimen for at least 4 wk before enrollment. Owners were informed that throughout the study period, the use of any new medications, supplements, dose changes, or other treatment strategies should be minimized and would have to be reported and would result in exclusion from the study.

Participants were allocated using an online randomizing software[a] to one of the following two groups: placebo followed by CBD treatment (PL-CBD) or CBD followed by placebo treatment (CBD-PL). After a 4 wk period for baseline measurements, either CBD or placebo treatment was initiated for 6 wk depending on the patient's group assignment. After receiving the first treatment for 6 wk, animals were crossed over to the opposite treatment for the subsequent 6 wk. Given the short (<4 hr) half-life of the product, the carry-over effect was considered negligible.[22] Therefore, no washout was instituted between treatment periods.

The company providing the CBD oil[b] dispensed two identical bottles to the research team. Both bottles contained cold-pressed hemp seed oil (confirmed via testing on Agilent high-performance liquid chromatography with a diode-array detector instruments using validated methods) and chicken flavoring as their base in an attempt to disguise the smell of the CBD. The CBD oil contained a highly purified CBD product in addition to the hemp seed oil and flavoring. The CBD product was made from a cannabis plant that was certified by the Colorado Department of Agriculture to contain <0.3% THC by dry weight. The plant-derived CBD oil was composed predominantly of CBD but also included small amounts of other cannabinoids, including THC, cannabidiolic acid, cannabinol and cannabigerol. The CBD oil was dosed at 2.5 mg/kg CBD *per os q* 12 hr, and the placebo was administered in equivalent volumes. The CBD or placebo oil was then provided to the owners in individual bottles containing identical information and dosing instructions for each patient. The owners and all personnel involved with the study were blinded to the contents of each bottle.

The following outcome measures were collected at the outlined time points throughout the study (**Figure 1**).

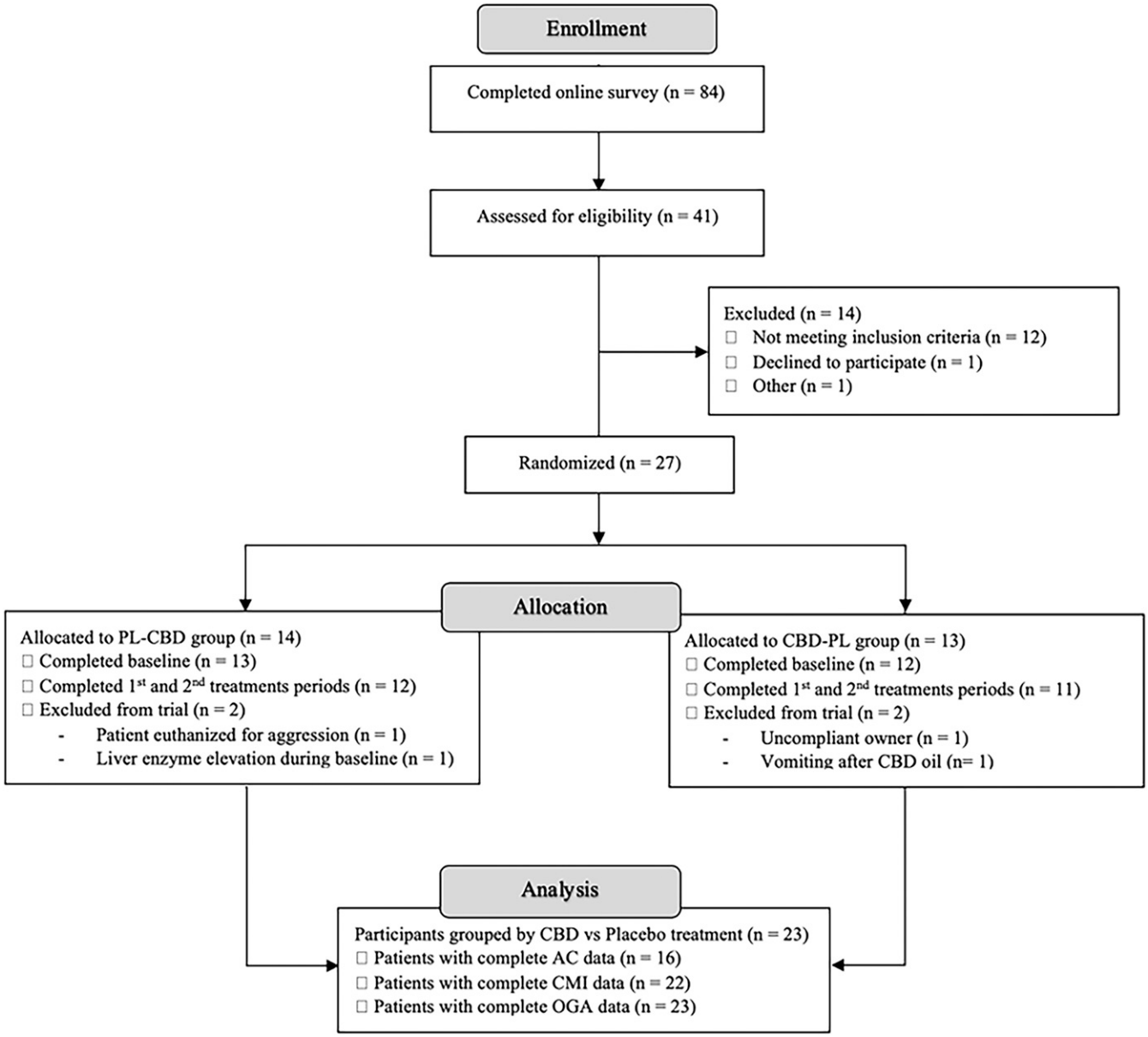

FIGURE 1

Number of participants for each phase of the clinical trial, from enrollment to data analysis. AC, activity count; CBD, cannabidiol; CBD-PL, CBD followed by placebo treatment; CMI, clinical metrology instrument; OGA, objective gait analysis; PL-CBD, placebo followed by CBD treatment.

Clinical Pathology

Elevations in liver enzyme levels were classified as either mild (greater than or equal to twofold and less than or equal to sixfold) or moderate (greater than sixfold).[23] Plasma CBD levels were measured at the crossover point and at the end of the study using a validated liquid chromatography–mass spectrometry-based assay. Briefly, 50 μL of unknown, standard, or quality control sample was added to a 1.5 mL microcentrifuge tube and 5 μL of 100 ng/mL d3CBD (deuterated internal standard) followed by 50 μL acetonitrile. Samples were mixed and centrifuged for 5 and 10 min, respectively. The resulting supernatant was transferred to an autosampler vial for analysis. Quantification was done using summed values of the measured transitions for CBD corrected for d3CBD. Assay performance was linear from the lower limit of quantitation of 1 ng/mL to 10 ug/mL and quality control samples showed an accuracy and precision of 93.1 and 4.6%, respectively.

Clinical Metrology Instruments

CMIs (Liverpool Osteoarthritis in Dogs and CBPI) were completed by the owners between weeks 1 and 4 during the baseline period, at week 10 (crossover point), and at week 16 (end of the study). All initial CMIs were discussed with the owner by the same investigator (S.M.), and all subsequent CMIs were completed by the same owner via dependent interviewing. Only complete CMIs were included in the analysis.

Accelerometry

Total activity counts (ACs) were measured using at least one of two different devices (Actical or Heyrex)[c,d]. To derive similar activity data from both devices, the percentage of change in total AC of the means from weeks 1 to 4 (baseline) and weeks 1 to 3 and 4 to 6 for each of the two treatment periods was used for comparison between groups. One of the accelerometers[c] was attached by removing the metal ring on the collar used for leash attachment and securing the accelerometer with two zip ties, whereas the second device[d] was attached next to it using the attachment provided by the company.[24] The monitors were placed in the same position relative to one another and were positioned ventral to the mandible of each dog. Data were recorded continuously throughout the study period. The epoch or window length for both devices were set to 60 s. Only data sets with at least 140 min of recorded activity for each day of the study duration were used for analysis.

Objective Gait Analysis

Gait analysis was performed once weekly during the baseline period (weeks 1 to 4), and then every 3 wk after initiation of the first treatment period using a pressure-sensitive walkway (PSW)[e]. Dogs were evaluated at a trot in a similar fashion to a previously described protocol.[25] If dogs were unable to trot, they were evaluated at a walk. All dogs were acclimated to the gait analysis laboratory and lead-walking on the left and right before their first pass until they showed no signs of anxiety. The first two trials were discarded. Six trials (three in each direction) with a subjectively constant velocity, in a straight line, without lateralization of the head, pulling on the lead, or stepping off of the PSW were acquired. When dogs only tolerated to be walked in one direction, then six trials in that single direction were acquired. The dogs were trotted over the PSW at their own comfortable speed within a velocity range of 1.7–2.2 m/s for the trot and 0.8–1.4 m/s for the walk. Trials from the subsequent visits for an individual dog were only considered valid if they fell within a velocity range of 0.3 m/s of their velocity established at week 1. The labeling of each foot placement by the program was visually confirmed by review of the video recorded during the gait analysis data acquisition. The following parameters were calculated and averaged from the six valid trials of each visit for each limb: peak vertical force normalized by body weight (PVF%) and percentage of body weight distribution (%BWD). These were calculated using the following formulas:

$$\text{PVF\%} = \text{PVF [N]}/(\text{BW [kg]} \times 9.8066)$$

$$\text{\%BWD} = (\text{PVF [N] of the limb/total PVF [N] of all four limbs in one gait cycle}) \times 100$$

Statistical Analysis

For statistical analysis, the initial established groups based on treatment sequences (i.e., CBD-PL and PL-CBD) were converted to treatment groups (i.e., CBD versus placebo) by grouping the values of all outcome measures after CBD and placebo treatments, respectively. Continuous outcome data were presented using mean (±standard deviation) and evaluated for normality (using Shapiro-Wilk Statistic and Q-Q Plot) before performing linear regression analysis to compare treatment groups within time points as well as time points within each treatment. All analyses were adjusted for the within-subject factor (i.e., same subject evaluated at multiple time points). The CMI score data were presented using medians and analyzed using a signed rank test for paired data to compare scores between the time points as well as between treatment groups. A P value of $\leq .05$ was used to determine statistical significance. All data were analyzed using commercially available software.[6]

Results

The number of dogs surveyed, evaluated, enrolled and included for analysis is summarized in **Figure 2**. Twenty-three dogs completed the study resulting in 11 participants in group PL-CBD and 12 in

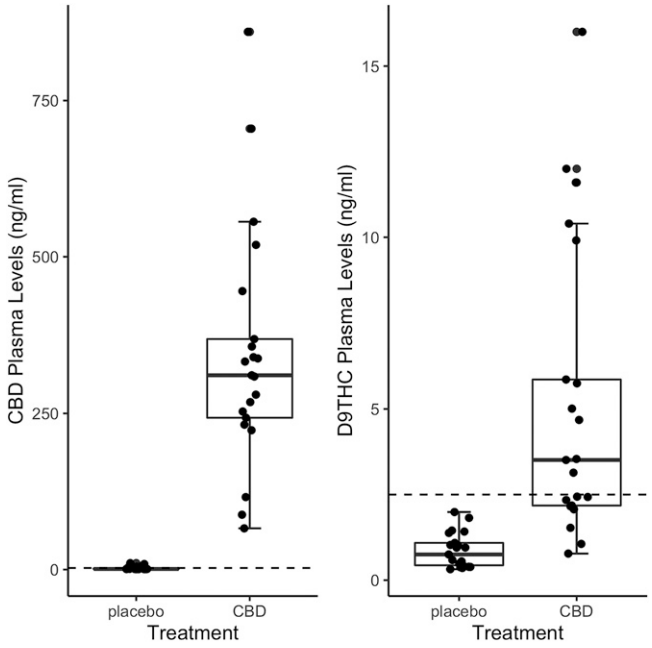

FIGURE 2

Box plot comparing median CBD and D9THC plasma levels (ng/mL) after 6 wk of treatment with either placebo or CBD oil. The dotted line represents the LLOQ. CBD, cannabidiol; D9THC, delta-9-tetrahydrocannabinol; LLOQ, lower limit of quantification.

group CBD-PL. Eleven dogs (48%) were spayed females, and 12 (52%) were neutered males. Age ranged from 4 to 14 yr (median 10 yr), and weight ranged from 22 to 63 kg (median 33 kg). Breeds and clinically affected joints are summarized in **Table 1**.

There were no differences noted between treatment groups at any time point for any of the recorded outcome measures. A statistically significant difference (i.e., a decrease of CMI scores and increase in ground reaction forces) between time points within treatment groups was observed for the following comparisons for the CBD group: CBPI Pain Severity Score, CBPI PIS, Liverpool Osteoarthritis in Dogs, and %BWD at weeks 3 and 6. For the placebo group, a significant difference was only observed for CBPI PIS. A decrease in AC from baseline was observed for both groups (3.56 and 9.96% at week 6 for CBD and placebo treatments, respectively); however, no statistically significantly difference was observed for either group. The CMI and ground reaction force data are detailed in **Tables 2** and **3**, respectively.

A post hoc sample size calculation was performed using SAS Proc Power[f] for a crossover design with alpha = 0.05. The baseline data were used to determine variability for %BWD of the affected limb (baseline average was 21.59) and the standard deviation (of differences after versus before treatment) was 1.45. Sample size calculation was performed for 80% power. To detect a 2.5 or 5% increase from baseline, a sample size of n = 59, or n = 17 animals, was determined.

Clinical Pathology and Adverse Effects

Fourteen dogs displayed mild elevations in liver enzymes after initiating treatment with CBD oil. One dog experienced liver enzyme elevation during baseline; no elevations in liver enzymes were observed after placebo treatment. Thirteen dogs (56%) received concomitant NSAID therapy throughout the study period, eight of whom displayed liver enzyme elevations during CBD administration. None of these dogs displayed clinical symptoms, and concomitant treatment was continued without further owner-reported adverse events. For the dogs with liver enzyme elevations that had available recheck serum chemistry 6 wk after CBD administration (i.e., CBD-PL group; n = 8), alkaline phosphatase values decreased to within the reference interval in five dogs and <500 IU/L in the remaining three dogs.

Two dogs vomited after CBD oil treatment was initiated: one dog was excluded because of intolerance of the CBD oil treatment (vomiting); this dog was not included in the data analysis. The other dog had mild vomiting during the first few days of CBD treatment, which resolved without any modification to the treatment regimen.

Adverse effects associated with CBD oil administration and individual plasma CBD concentrations measured at the end of the CBD administration period are presented in Table 1.

CBD Levels

The median CBD plasma levels after 6 wk of CBD oil were 311 ng/mL (range 5–860), and the median CBD plasma levels after 6 wk of placebo were 0.96 ng/mL (range 0.6–572; Figure 2).

Discussion

This study was conducted to acquire further pilot data to determine the clinical effect of CBD on canine OA-associated pain in client-owned dogs using objective outcome measures. At the time of writing, only one relevant paper has evaluated the clinical question of whether CBD provides additive pain control in dogs with OA,[26] and to our knowledge, this is the first study to report objective gait analysis data. Given the small sample size, no definitive conclusions can be drawn from the data; however, the provided results clearly indicate that further research is needed before recommending CBD for clinical use. Similarly, the observed adverse events (vomiting and liver enzyme elevation) also warrant further investigation, particularly because long-term administration of the product is anticipated when used in patients with OA.

The elevation in liver enzymes in the present study is consistent with previous reports in dogs and humans.[27] Liver enzyme elevations

TABLE 1

Summary of All Enrolled Patients, Clinical Diagnoses of Affected Joints, and CBD Plasma Levels (for Dogs Included in Final Analysis) after 6 wk of CBD Oil Administration

Patient No.	Breed	Affected Joints	Sex	Age, yr	NSAID	Adverse Effects Associated with CBD Administration	CBD Plasma Levels, ng/mL
1	Labrador retriever	Bilateral elbow	CM	12	Carprofen	ALP 160; ALT 243; AST 50	66
2	Pit bull terrier	L elbow	CM	11	Carprofen	—	311
3	Collie	>3 joints	SF	10	Carprofen	ALP 378; ALT 161	750
4	Staffordshire bull terrier	Bilateral elbow	SF	10	No NSAID	ALP 549; ALT 94; AST 46	338
5	Mixed-breed dog	Bilateral stifle	SF	13	Meloxicam	ALP 1455; ALT 121; AST 281	705
6	German shepherd dog	Bilateral stifle	M	10	Carprofen	ALP 241	268
7	Labrador retriever	Bilateral elbow	SF	7	Grapiprant	—	333
8	Mixed-breed dog	R elbow	SF	8	Carprofen	ALP 1016	369
9	Bulldog	R elbow	SF	8	No NSAID	ALP 157	243
10	Border collie	L elbow	CM	13	No NSAID	—	5
11	Staffordshire bull terrier	Bilateral elbow	SF	10	No NSAID	—	88
12	Newfoundland	Bilateral elbow	SF	4	No NSAID	—	309
13	Labrador retriever	Bilateral hip	SF	8	No NSAID	ALP 2493; Vomiting	232
14	Mixed-breed dog	Bilateral elbow and L stifle	CM	12	No NSAID	ALP 1134; ALT 131	357
15	Pit bull terrier	Bilateral stifle	CM	10	Carprofen	ALP 702; ALT 247	556
16	Boxer	Bilateral stifle	CM	11	No NSAID	—	519
17	German shepherd dog	Bilateral hip	CM	9	No NSAID	ALP 224	116
18	Mixed-breed dog	Bilateral elbow	SF	14	Carprofen	—	280
19	Great Pyrenees	L hip and R shoulder	FS	8	No NSAID	ALP 182	445
21	Entlebucher	R elbow	MN	9	Meloxicam	ALP 263	223
22	Labrador retriever	Bilateral hip	MN	6	No NSAID	—	860
23	Labrador retriever	Bilateral elbow and hip	MN	12	Carprofen	—	340
24	Labrador retriever	Bilateral elbow	MN	9	Carprofen	ALP 3150	253

Reference range for liver enzymes: ALP, 15–140 IU/L; ALT, 10–90 IU/L; AST, 15–45 IU/L.
—, no adverse effects; ALP, alkaline phosphatase; ALT alanine aminotransferase; AST, aspartate aminotransferase; CBD, cannabidiol; CM, castrated male; L, left; M, male; NSAID, nonsteroidal anti-inflammatory drug; R, right; SF, spayed female.

associated with CBD administration may be due to an induction of the cytochrome-P450–mediated oxidative metabolism of the liver.[18,28] Unfortunately, the hepatotoxic potential of CBD cannot be determined from this study. However, previous research suggested a lack of liver dysfunction when liver enzyme elevation is observed after CBD administration in healthy dogs.[27] In that study, 30 dogs were administered up to fourfold the dose of the same product used in the current study for 6 wk. Elevation of alkaline phosphatase was observed in 36% (n = 11) of the dogs, whereas alanine aminotransferase, aspartate aminotransferase, and pre- and postprandial bile acids remained normal in all dogs throughout the study period. Although liver enzyme elevations may simply be due to enzyme induction, further investigation into its impact on hepatic architecture and clinical relevance is warranted.

Concomitant NSAID administration may play a role in the observed enzyme elevation. Although CBD interactions with other medications have been previously reported,[28, 29] its interaction with NSAIDs has not been studied in dogs. NSAIDs can be associated with liver enzyme elevation in dogs.[6] However, six dogs that were not receiving NSAIDs in this study also displayed liver enzyme elevation. As previously suggested,[27] further studies investigating the clinical implications of long-term administration of CBD and drug interactions between CBD and NSAIDs are necessary, particularly because combined administration would be desirable to allow for multimodal pain management.

The other adverse event, vomiting, was observed in less than 10% of the dogs, and the remaining participants tolerated CBD treatment well with no owner-reported side effects. Because this

TABLE 2

Baseline and Post-Treatment Median (±SD) Scores and Associated *P* Values for the CMI Data (CBPI and LOAD) Comparing Time Points Within Each Treatment Group (CBD and Placebo) and Between Treatment Groups

CMI	Treatment Group	Baseline CMI Score, Median ± SD	Post-Treatment CMI Score, Median ± SD	*P* Value Comparing Time Points Within Treatment Groups	*P* Value Comparing Time Points Between Treatment Groups
CBPI PSS (0–40)	CBD	17.64 ± 6.37	14.73 ± 7.08	.018*	.89
	Placebo		14.86 ± 5.74	.093	
CBPI PIS (0–60)	CBD	32.76 ± 11.80	26.71 ± 13.12	.016*	.59
	Placebo		24.81 ± 12.91	.007*	
LOAD (0–52)	CBD	28 ± 6.88	24.91 ± 8.05	.019*	.74
	Placebo		25.05 ± 8.48	.09	

*Indicates statistically significant values ($P \leq .05$)
CBD, cannabidiol; CBPI, Canine Brief Pain Inventory; CMI, clinical metrology instrument; LOAD, Liverpool Osteoarthritis in Dogs; PIS, pain interference score; PSS, pain severity score; SD, standard deviation.

adverse event was only observed infrequently, it is possible that it was not associated with administration of CBD.

Recent randomized placebo-controlled and double-blinded studies using CMIs and veterinary exams as outcome measures concluded that CBD may benefit dogs with OA.[18,20] Although Gamble et al.[18] did not report a statistically significant caregiver placebo effect (this could be related to the odor differences between treatments or, alternatively, as suggested by the authors, be associated with the fact that a lot of the participants were in the medical field), our study found a significant improvement for one of the CMIs used (PIS) after both CBD and placebo treatment. The observed improvement in the control group may be attributed to either a caregiver placebo effect (which has been reported to occur up to 57% of the time when owners evaluate their dog's lameness[30]) or a potential benefit of the hemp seed oil base used for the control group. A significant improvement was also detected for comparisons between time points within the CBD treatment group for one of the ground reaction force measurements (%BWD). However, no difference was observed for the same comparison for PVF%. As such, the observed difference for %BWD may be attributed to either a Type I statistical error or a true difference. Obviously, a simultaneous increase in PVF% would more clearly support a true difference. However, Kano et al. have recently shown that %BWD is the most accurate gait parameter when evaluating a heterogeneous group at a controlled velocity: %BWD was associated with a lower variability than PVF % in their study.[31] It is important to note that we did not detect a statistically significant difference for comparisons of %BWD or PVF% between time points within the placebo treatment group or for any of the comparisons between treatment groups. Our post hoc sample size calculation suggests that we would have identified a difference between treatment groups if the differences in %BWD would have exceeded 5% (calculated minimum sample size of n = 17). Yet, a difference of ≥2.5% may not have been identified with

TABLE 3

Mean (±SD) Data and Associated *P* Values of Ground Reaction Forces Comparing Time Points (i.e., Baseline to W3/W6) Within Each Treatment Group (CBD and Placebo) and Between Treatment Groups

Ground Reaction Forces	Treatment	Baseline Mean ± SD	W3 Mean ± SD	*P* Value Comparing Time Points Within Treatment Groups	*P* Value Comparing Time Points Between Treatment Groups	W6 Mean ± SD	*P* Value Comparing Time Points Within Treatment Groups	*P* Value Comparing Time Points Between Treatment Groups
PVF%	CBD	52.32 ± 16.95	54.25 ± 19.97	.085	.735	53.86 ± 19.11	.15	.77
	Placebo		54.63 ± 20.39	.1377		53.63 ± 18.69	.23	
%BWD	CBD	21.59 ± 3.81	22.47 ± 4.43	.0013*	.24	22.17 ± 4.03	.05*	.73
	Placebo		22.07 ± 4.38	.22		22.07 ± 4.37	.16	

*Indicates statistically significant values ($P \leq .05$).
%BWD, percentage of body weight distribution; CBD, cannabidiol; PVF%, peak vertical force normalized by body weight; SD, standard deviation; W3, Week 3; W6, Week 6.

the sample size of the present study (calculated minimum sample size of n = 59).

The observed plasma CBD concentrations described in this study are higher than previously reported values,[18] which may be due to the slightly higher dose (2.5 mg/kg versus 2 mg/kg) or a difference between products. An early pharmacokinetics study reported that CBD in dogs yielded a low bioavailability after oral administration of a single dose of 180 mg (7.5–11.25 mg/kg) of CBD. However, that particular study did not report half-life values after oral administration.[32] Furthermore, that study investigated a powder-based product as opposed to the oil-based preparations used in the present and another recent study.[18] One dog in the present study showed a low CBD level (5 ng/mL) after CBD treatment, which may be due to a lack of absorption.[34] However, the remainder of the dogs had measurable CBD levels (66–860 ng/mL) after CBD treatment. The broad distribution in CBD levels may warrant testing of CBD levels for each individual dog to ensure proper absorption. One dog showed a high CBD level (572 ng/mL) after placebo treatment, for unknown reasons.

Our study has several limitations, including the short duration of data collection, small sample size, use of a novel accelerometer[d], lack of a washout period, potential difficulties with the blinding process, use of hemp oil as the base for the placebo group, and inconsistent NSAID use.

A novel accelerometer device was initially used to allow for remote data download via Wi-Fi. However, after technical difficulties resulted in data loss, study participants were also equipped with a conventional accelerometer[c]. To allow for analysis of AC using the two devices, a change (i.e., increase or decrease) in weekly AC was used, which may have affected our results. We observed a decrease in AC from baseline after treatment with both CBD oil and placebo. It is unclear why the AC data measured in this study revealed a decrease from baseline values after CBD and placebo treatments. However, these changes fall within the range of the normal expected variation, because an increase of 20% has been suggested to be clinically relevant for dogs receiving NSAID therapy.[33] It is worth noting that accelerometry has a questionable value as an outcome measure in research related to canine activity because the displayed data tend to be more a reflection of owner behavior (i.e., increase or decrease in exercise or outdoor activities) rather than an actual increase in comfort (or decrease in pain) from the patient. However, Brown et al. described a significant increase in activity counts when comparing dogs with naturally occurring OA treated with carprofen versus dogs treated with placebo.[33]

A general recommendation for pharmacological studies is that the washout period should be approximately five times the half-life of the product tested (to avoid a carry-over effect).[34] Carry-over effect occurs when the treatment given over the first period of the study carries over into the second period (in this case, the CBD-PL group). This may lead to exaggerated responses during the placebo administration and, therefore, to an overall reduction in apparent effectiveness of the treatment.[35] A recent study with a similar study design used a 4 wk treatment period with a 2 wk washout period.[18] Given the short half-life of CBD, rather than including a washout period, we opted to collect outcome measures at multiple time points (i.e., at 3 and 6 wk after initiation of treatment or placebo) and adjust the analysis for the within-subject factor. This allowed a shortened study timeline (thereby reducing the risk of attrition), while assessing a slightly longer treatment period than in a previous study.[18] However, given the inconsistent CBD levels, the authors encourage future studies to be performed with a washout period of at least 1 wk.

Hemp seed oil was used as the base for both treatment groups to disguise the smell of the product. This may have resulted in a positive clinical effect due to the polyunsaturated fatty acids or even low levels of cannabinoids contained in the product.[36] The beneficial effects of polyunsaturated fatty acids have been well documented; however, the purpose of this study was to investigate the additional effects of CBD. Given the widespread use of polyunsaturated fatty acids for clinical treatment of OA in dogs, this is the most clinically relevant question. However, other studies have noted that a lack of difference between treatment groups may be due to a treatment effect of the placebo product.[37] Despite our efforts, we were unable to fully duplicate the strong characteristic smell of the CBD oil in our placebo product. This may have affected the CMI data, which should be considered when interpreting the results of the present study. In the authors' opinion, greater emphasis should be placed on objective outcome measures, such as objective gait analysis and accelerometry, because the CMI scores may be influenced by the owner's blinding status.

To allow for quicker enrollment, dogs were allowed to receive NSAID therapy throughout the study period if they had previously received this therapy consistently. Clearly, NSAID therapy is a confounding factor given its efficacy for treating pain, if used inconsistently throughout the study. However, our study was designed to eliminate NSAID use as a confounding factor because owners were asked to continue the same administration regimen throughout the study period (otherwise resulting in exclusion from data analysis). Therefore, the chosen crossover study design allows to evaluate the additive effects of CBD as long as NSAID administration is consistent throughout the study period. This holds true for any confounding factor (e.g., the use of hemp oil, nutraceuticals, other pain medications), which is why a consistent OA treatment regimen was one of the inclusion criteria for enrollment in the study. Alternative

approaches would be to either discontinue NSAIDs before initiation of the study or place all dogs on NSAIDs. Although these approaches represent the preferred study design, they either compromise pain control of participants or limit the number of possible patients that can be enrolled.

Conclusion

The pilot data from this study do not support the use of CBD as a symptom-relieving agent for canine OA. However, given the small sample size and conflicting results of previous research, further studies may be warranted. Future research should also assess the long-term safety of CBD administration for OA-associated pain as well as the possible interactions with NSAIDs.

FOOTNOTES

[a] Random.org; Randomness and Integrity Services, Ltd., Dublin, Ireland
[b] Applied Basic Science Corporation, Castle Rock, Colorado
[c] Actical; Philips Respironics, Murrysville, Pennsylvania
[d] Heyrex; Philips Respironics, Wellington, New Zealand
[e] Tekscan HRV Walkway 6 VersaTek system; Tekscan Inc, South Boston, Massachusetts
[f] SAS version 9.4; SAS Institute Inc., Cary, North Carolina

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