ENHANCING CANNABIDIOL BIOAVAILABILITY THROUGH A NATURAL PHYTOCHEMICAL ADDITIVE

J.T. Hastings and K.M. Schmidt*

Research & Development, Centuria Foods, Inc.,
Carson City, NV 89706 USA

ks@centuriafoods.com

Research at Centuria Foods has led to the development of several water-soluble formulations which increase the oral bioavailability of cannabidiol (CBD). Our C10-Clear and C10-Cloudy formulas show a marked improvement over traditional oil-based tinctures containing CBD isolate and extracts. In addition, incorporating our novel Boost process into the emulsifying stage of production leads to a 30-40% increase in overall CBD uptake, higher absorption rates, and sustained duration as compared to non-Boosted preparations.

1 Introduction

Cannabidiol (CBD) is a phytocannabinoid compound found within the Cannabis plant, which has garnered increased attention due to its therapeutic potential for a number of ailments. Thus far, CBD has shown promise in treating various conditions including epilepsy[1], Parkinson's disease[2] , and Alzheimer's disease, as well as in the management of pain, anxiety, and depression[3,4]. For those who use CBD as a treatment for these conditions, a high level of safety with few adverse side effects is observed[5]. Additionally, CBD is known for its anti-inflammatory[6], anti-seizure[7], and anxiolytic properties[8]. The added benefit of using CBD as a therapy is that it is nonpsychoactive, non-addictive, and has no intoxicating effects, unlike tetrahydrocannabinol (THC), another cannabinoid found in the Cannabis plant[9].

Cannabinoids can be consumed in a number of ways, with the most common forms of administration for pain relief including smoking and oral ingestion via cannabinoid-infused food, pills, and beverages. One of the major challenges regarding the use of CBD as a treatment is maximizing the uptake of orally-ingested CBD into the body for extended periods of time with little delay[10]. In comparison to intravenous injection or inhalation, there are some advantages to oral administration—it is not invasive as with intravenous medication and does not cause irritation to the throat or coughing as with inhalation. However, the bio-availability of CBD administered through oral ingestion is lower than other methods, and users often experience the effects after 30-90 minutes[11].

One possible way of increasing uptake via oral ingestion is infusing CBD into water. CBD and water are normally immiscible to one another due to the hydrophobic nature of CBD. Ultrasonic emulsification has been used by both the pharmaceutical[12] and food processing[13] industries to encapsulate hydrophobic compounds inside a surfactant shell to dissolve oil droplets into water without separation[14]. Encapsulating CBD into sub-micron droplets via ultrasonication does have potential to allow for more effective bioavailability as compared to ingesting CBD in an oil tincture form[15].

To address these issues, several C10 formulations were developed to provide enhanced bioavailability and a faster response time to experience therapeutic effects.

2 Materials & Methods

2.1 Sample Preparation

The oil tincture and isolate samples were prepared by dissolving the extracts in sunflower seed oil to obtain the desired CBD concentrations. The mixtures are then heated in a water bath until a single phase exists.

Samples of C10-Clear contain a proprietary blend of ingredients including water, hydrogenated castor oil, hemp oil concentrate, fractionated coconut oil, and preservatives. C10-Cloudy contains water, hemp oil concentrate, fractionated coconut oil, plant saponins, and preservatives. In the Boosted samples, the proprietary phytochemical additives displace an equivalent amount of fractionated coconut oil. After processing to create an emulsified mixture, these water-soluble materials are diluted with distilled water to produce the correct dosage levels.

2.2 Animals/Environment

Male rats (strain Crl:CD SD, Charles River Laboratories Japan) aged 7-8 weeks weighing between 200-300 grams were placed (groups of four) in a polymethylpentene animal cage having dimensions 292 mm x 440 mm x 200 mm (W×D×H). External conditions are set such that there is a 12-hour light/dark cycle (switch at 0600/1800), the average temperature was kept in the range 20–26°C, relative humidity was between 30% and 70%, they were exposed to the ALPHA-dri bedding material (Shepherd Specialty Papers Inc.), and each rat had free access to clean water (public supply) and fresh air. Rats were placed on a diet consisting of MF (Oriental Yeast Co., Ltd.). Prior to the study, the animal use protocol was reviewed and approved by the Institutional Animal Care and Use Committee of Sekisui Medical Co., Ltd. (SMD), and all animal studies were carried out according to these protocols.

Table 1: CBD plasma concentrations

Time (min)	Isolate	Oil Tincture	C10-Clear		C10-Cloudy	
			Boost	No Boost	Boost	No Boost
10	ND	ND	4.08±0.53	3.34±1.60	1.55±0.63	1.85±1.67
30	ND	ND	12.00±1.80	8.99±5.91	2.87±1.76	2.30±1.02
60	ND	ND	7.40±2.19	4.74±2.78	2.22±1.77	1.04±0.35
120	ND	0.19±0.20	2.14±0.61	1.60±0.70	1.02±0.52	0.85±0.77
240	0.28±0.31	2.04±1.18	0.85±0.15	0.66±0.11	1 0.42±0.09	0.49±0.14

Concentration of CBD in the blood (ng/μL) given as μ ±σ (n=3). ND: Non-Detect.

2.3 Pharmacokinetic Experiments

Starting at 24 hours prior to the trial, one rat was removed to leave three animals per cage and allow for each experiment to be run in triplicate. Fasting conditions were set starting at 16 hours prior to and 4 hours following administration of the CBD sample. Each animal was given a dose of 2 mg CBD / kg body weight from a stock solution at 0.4 mg CBD / mL (5 mL/kg total). A syringe fitted with an oral tube is used to administer the material into the stomach.

Approximately 300 mL of blood is collected from the jugular vein using a heparinized syringe with an injection needle. Following centrifugation (8000×g, 4°C, 5 min), the plasma is separated, collected in a polypropylene tube, and stored in a freezer to await analysis by LC-MS/MS. Five time-points are sampled for each rat at: 10 min, 30 min, 1 hr, 2 hr, and 4 hr following drug administration.

3 Results and Discussion

Table 1 lists the blood plasma levels of CBD taken at various times throughout the experiment. Absorption of cannabinoids from the oil phases has a significantly delayed onset time in addition to large values of T_{max}, the time required to reach maximum concentration (C_{max}). For the oil tincture and isolate, it takes approximately 2 and 4 hours, respectively, before the concentration of CBD in blood plasma reaches a detectable value. This corresponds to a drastic lag-time for consumers utilizing CBD before they are able to experience the therapeutic effects and can confirm a proper dosage has been taken.

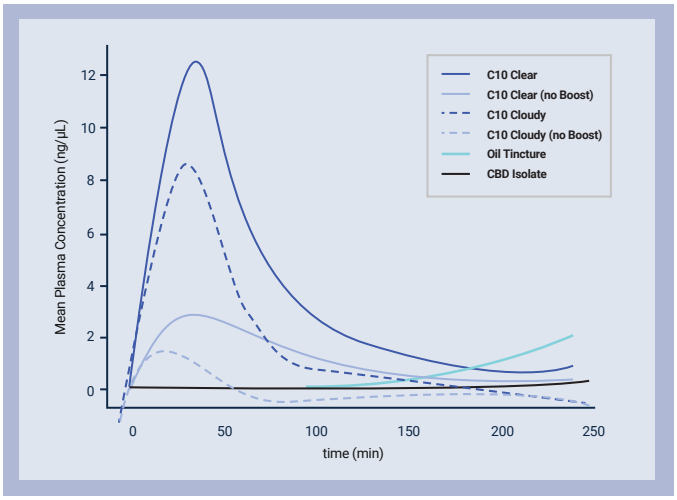

Figure 1: Plasma concentration-time curves of cannabidiol in various formats administered orally to rats at a dosage of 2 mg CBD / kg body weight.

Each of the water-soluble C10 formats show a significant reduction in the absorption time compared to CBD isolate and the oil tincture. The concentration of CBD reaches detectable values after only ten minutes following oral ingestion of C10 and surpasses the maximum concentrations observed for the oil-soluble formats found after four hours. This effect is graphically represented in **Figure 1**, which displays the plasma concentration-time curves for each of the materials used. All emulsions allow for quick absorption of CBD into the blood stream and reach maximum concentrations after 30 minutes. In addition, the concentration observed after 30 minutes by C10-Clear and C10-Cloudy are 5.88 and 1.41 times greater than that obtained at 4 hours with the oil tincture, respectively.

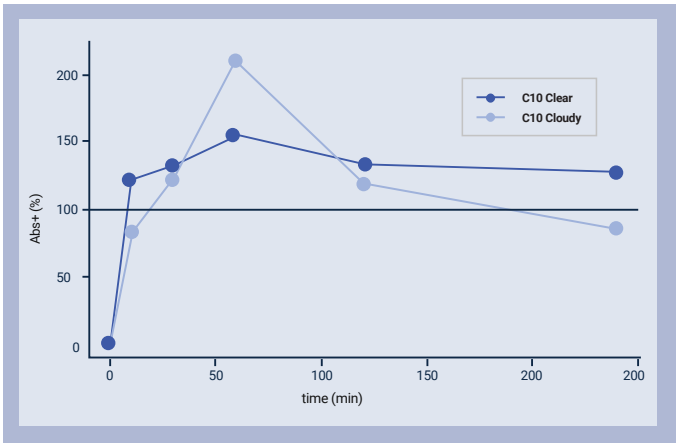

Figure 2: Effect of Boost formula on bioavailability of C10-Clear and C10-Cloudy emulsions. Absorption enhancement, Abs+(t), is calculated using Eqn. (1).

The effect of our Boost formulation on bioavailability is shown in **Figure 2**, where absorption enhancement is calculated by the ratio of plasma concentrations of C10 formulations containing Boost (C_{Boost}) to those without Boost ($C_{no\ Boost}$):

$$Abs^{+}(t) = \frac{C_{Boost}(t)}{C_{no\ Boost}(t)}$$

Parameter	Isolate	Oil Tincture	C10-Clear		C10-Cloudy	
			Boost	No Boost	Boost	No Boost
C_{max} (µg/mL)	0.28	2.04	12.00	8.99	2.87	2.30
T_{max} (min)	240	240	30	30	30	30
$AUC_{(0-10)}$(mg · min/L)	0.00	0.00	20.40	16.70	7.75	9.25
$AUC_{(0-\infty)}$(mg · min/L)	16.68	139.32	937.98	671.87	311.96	238.07

Cmax: peak plasma concentration; T_{max}: time to reach C_{max}; $AUC_{(0-10)}$: integrated value of concentration-time curve after 10 minutes; $AUC_{(0-\infty)}$: integrated value of concentration-time curve after final data point.

Table 2: Pharmacokinetic parameters from plasma concentration-time curve

Both Boost formulas showed a marked improvement over the non-Boost counterparts. At all recorded intervals, CBD levels are C10-Clear are increased by at least 20% with the addition of Boost. C10-Cloudy has a 20% enhancement between 10 and 120 minutes following ingestion, more than doubling the plasma concentrations at one hour when compared to non-Boosted C10-Cloudy. This efficiency gain correlates to an increase in the overall amount of CBD absorbed and increased effective time.

Several parameters exist to aid the evaluation of pharmacokinetic studies to assess the rate and extent of drug absorption[16]. These include the peak plasma concentration (C_{max}), time required to reach this concentration (T_{max}), and the integral of the concentration-time curve up to some time 't' ($AUC_{(0-t)}$), and are provided in **Table 2**. Information on the dynamics of absorption can be inferred from C_{max}, such that elevated C_{max} values are associated with increased absorption rate[16]. Both C10 formulations result in higher C_{max} values compared to the oil-based products, with significantly higher concentrations shown for C10-Clear.

The area under the concentration-time curve ($AUC_{(0-t)}$) allows one to quantify the extent of absorption for a given dosage by summing up the concentration levels throughout the entire duration of the experiment. Calculated values of AUC are given in **Table 2** at 10 minutes and 4 hours following administration of the samples. The C10-Clear formulas (Boost and non-Boost) give similar values and nearly double those shown by C10-Cloudy, though C10-Clear has a 22% increase over its non-Boosted counterpart. The 4-hour integrations, given by $AUC_{(0-240)}$, provide an estimate of the total absorption of each sample. The oil-based routes lead to a considerable reduction in total absorption when compared to C10. In fact, the 10-minute $AUC_{(0-10)}$ values for both C10-Clear emulsions surpass the final $AUC_{(0-240)}$ for CBD isolate.

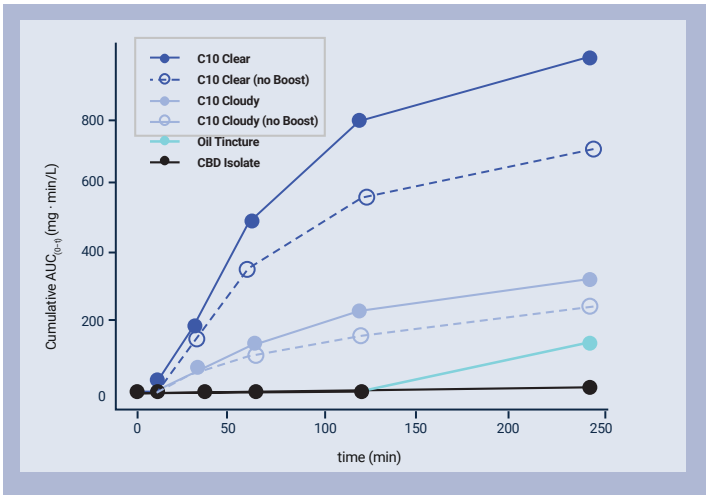

Figure 3: Cumulative values of $AUC_{(0-t)}$ to quantify the extent of absorption for each format used in this study. $AUC_{(0-t)}$: area under the curve calculated using the trapezoidal rule.

To better visualize these results, cumulative $AUC_{(0-t)}$ are presented in **Figure 3**, showing the total integration at each sampled time-point. All water-soluble dosage forms are seen to result in heightened levels of total CBD absorption. Additionally, incorporating Boost into the emulsions enhance the uptake levels even further. We can quantify these improvements with the relative bio-availability, $F^{B/A}_{rel}$, defined by:

$$F^{B/A}_{rel} = \frac{AUC_{B,(0-\infty)}}{AUC_{A,(0-\infty)}} \times \frac{D_A}{D_B}$$

where $AUC_{x;(0-\infty)}$ and Dx are the area under the concentration-time curve and dosage amount, respectively, for some solution 'x', and $F^{B/A}_{rel}$ gives the relative bioavailability of solution 'B' with respect to solution 'A'. Comparing the C10 ('B') to the non-Boosted emulsion ('A'), relative bioavailabilities at t = 240 are 1.40 and 1.31 for C10-Clear and C10-Cloudy, respectively.

Summary

As illustrated in this paper, Centuria Foods has engineered a synergy between a proprietary natural additive and emulsion technology to enhance the bioavailability and bioeffectiveness of CBD. Robustness of the natural additive and processing was shown by its increase in CBD uptake by more than 30% in both emulsion systems: C10-Clear and C10-Cloudy.

References

[1] Franco, V.; Perucca, E. Pharmacological and Therapeutic Properties of Cannabidiol for Epilepsy. *Drugs* **2019**, 79, 1435–1 1454.

[2] Zuardi, A. W.; Crippa, J.; Hallak, J. E. C.; Pinto, J.; Chagas, M. H. N.; Rodrigues, G.; Dursun, S.; Tumas, V. Cannabidiol for the treatment of psychosis in Parkinson's disease. *Journal of Psychopharmacology* **2009**, 23, 979–983.

[3] Hughes, B.; Herron, C. E. Cannabidiol Reverses Deficits in Hippocampal LTP in a Model of Alzheimer's Disease. *Neurochemical Research* **2019**, 44, 703–713.

[4] Philpott, H. T.; O'Brien, M.; McDougall, J. J. Attenuation of Early Phase Inflammation by Cannabidiol Prevents Pain and Nerve Damage in Rat Osteoarthritis. *Pain* **2017**, 158, 2442.

[5] Machado Bergamaschi, M.; Helena Costa Queiroz, R.;Waldo Zuardi, A.; Crippa, A. S., et al. Safety and side effects of cannabidiol, a Cannabis sativa constituent. *Current drug safety* **2011**, 6, 237–249.

[6] Petrosino, S.; Verde, R.; Vaia, M.; Allarà, M.; Iuvone, T.; Di Marzo, V. Anti-inflammatory properties of cannabidiol, a nonpsychotropic cannabinoid, in experimental allergic contact dermatitis. *Journal of Pharmacology and Experimental Therapeutics* **2018**, 365, 652–663.

[7] Jones, N. A.; Hill, A. J.; Smith, I.; Bevan, S. A.; Williams, C. M.; Whalley, B. J.; Stephens, G. J. Cannabidiol displays antiepileptiform and antiseizure properties in vitro and in vivo. *Journal of Pharmacology and Experimental Therapeutics* **2010**, 332, 569–577.

[8] Almeida, V.; Levin, R.; Peres, F. F.; Niigaki, S. T.; Calzavara, M. B.; Zuardi, A. W.; Hallak, J. E.; Crippa, J. A.; Abílio, V. C. Cannabidiol exhibits anxiolytic but not antipsychotic property evaluated in the social interaction test. *Progress in Neuro-Psychopharmacology and Biological Psychiatry* **2013,** 41, 30–35.

[9] Pavlovic, R.; Nenna, G.; Calvi, L.; Panseri, S.; Borgonovo, G.; Giupponi, L.; Cannazza, G.;
Giorgi, A. Quality traits of "cannabidiol oils": cannabinoids content, terpene fingerprint and
oxidation stability of European commercially available preparations. *Molecules* **2018**, 23, 1230.

[10] Mechoulam, R.; Parker, L. A.; Gallily, R. Cannabidiol: an overview of some pharmacological aspects. *The Journal of Clinical Pharmacology* **2002**, 42, 11S–19S.

[11] Grotenhermen, F. Pharmacokinetics and pharmacodynamics of cannabinoids. *Clinical pharmacokinetics* **2003**, 42, 327–360.

[12] Aulton, M. E.; Taylor, K. M. *Aulton's Pharmaceutics E-Book: The Design and Manufacture of Medicines*; Elsevier Health Sciences, **2017**.

[13] Loi, C. C.; Eyres, G. T.; Birch, E. J. In *Encyclopedia of Food Chemistry*; Melton, L.,
Shahidi, F., Varelis, P., Eds.; Academic Press: Oxford, **2019**; pp 404 – 409.

[14] Yaqoob Khan, A.; Talegaonkar, S.; Iqbal, Z.; Jalees Ahmed, F.; Krishan Khar, R. Multiple emulsions: an overview. *Current drug delivery* **2006**, 3, 429–443.

[15] Nakano, Y.; Tajima, M.; Sugiyama, E.; Sato, V. H.; Sato, H. Development of a novel nanoemulsion formulation to improve intestinal absorption of cannabidiol. *Medical Cannabis and Cannabinoids* **2019**, 2, 35–42.

[16] Urso, R.; Blardi, P.; Giorgi, G. A short introduction to pharmacokinetics. *European review for medical and pharmacological sciences* **2002**, 6, 33–44.