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Title Page

THE PHARMACOLOGICAL CASE FOR CANNABIGEROL (CBG)

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2. Running Title: Potential Clinical Uses of CBG

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3. Abstract

Medical cannabis and individual cannabinoids, such as tetrahydrocannabinol (Δ9-THC) and cannabidiol (CBD), are receiving growing attention in both the media and the scientific literature. The *Cannabis* plant, however, produces over 100 different cannabinoids, and cannabigerol (CBG) serves as the precursor molecule for the most abundant phytocannabinoids. CBG exhibits affinity and activity characteristics between Δ9-THC and CBD at the cannabinoid receptors, but appears to be unique in its interactions with alpha-2 adrenoceptors and 5-HT_{1A}. Studies indicate that CBG may have therapeutic potential in treating neurological disorders (e.g., Huntington's disease, Parkinson's disease, and multiple sclerosis), inflammatory bowel disease, as well as having antibacterial activity. There is growing interest in the commercial use of this unregulated phytocannabinoid. This review focuses on the unique pharmacology of CBG, our current knowledge of its possible therapeutic utility, and its potential toxicological hazards.

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4. Significance Statement

Cannabigerol (CBG) is currently being marketed as a dietary supplement and, as with cannabidiol (CBD) before, many claims are being made about its benefits. Unlike CBD, however, little research has been performed on this unregulated molecule, and much of what is known warrants further investigation to identify potential areas of therapeutic uses and hazards.

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6. Introduction

Cannabinoids as medicine

The use of *Cannabis sativa* as a medicine dates back millennia. In ancient China, marijuana was used to treat gout, malaria, digestive disorders, and menstrual pain (Bostwick, 2012; Russo, 2016; Kinghorn et al., 2017; Ryz et al., 2017; Baron, 2018; Ambrose and Simmons, 2019). Cannabis was introduced to western medicine by William O'Shaugnessy, who proposed its use for the treatment of rheumatism and seizures (Bostwick, 2012; Russo, 2016; Kinghorn et al., 2017; Baron, 2018). However, the use of medicinal marijuana fell out of favor towards the end of the nineteenth century and continued to decline until it was banned by the Controlled Substances Act of 1970 (Bostwick, 2012; Sacco, 2014; Kinghorn et al., 2017). Since that time, few cannabinoid drugs have been approved for human use. These drugs include: nabilone in 1985, dronabinol in 1986, rimonabant in 2006 (in Europe; withdrawn in 2008), Sativex (in Europe) in 2010, and Epidiolex in 2018. A growing number of countries have also approved the use of medical marijuana for treating a variety of medical conditions; however, the data that support the use of marijuana for treatment is often anecdotal or from small studies.

Cannabigerol and cannabinoid synthesis

Cannabinoid synthesis begins with the precursor molecules olivetolic acid and geranyl-pyrophosphate, which combine to form cannabigerolic acid (CBGA) (Shoyama et al., 1975; Fellermeier and Zenk, 1998; Fellermeier et al., 2001; Gülck and Møller, 2020). CBGA serves as the precursor to most other cannabinoids and is converted to Δ9-THCA (tetrahydrocannabinolic acid), CBDA (cannabidiolic acid), and CBCA (cannabichromenic acid) (Figure 1). Because CBGA serves as the precursory molecule to the other cannabinoids, it is normally found in very

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low quantities in *Cannabis*; however, strains with reduced activity of the three major synthesis enzymes can accumulate higher levels of CBGA (Fellermeier and Zenk, 1998; Fellermeier et al., 2001). All enzymatically produced cannabinoids (including CBG) are produced as their acidic form and are then decarboxylated by heat to create the "active" form.

With the recent de-regulation of cannabidiol (CBD) and other hemp-derived cannabinoids, such as CBG, cannabichromene (CBC), and cannabinol (CBN) (2018 Farm Bill), there is growing interest in cannabinoid pharmacology (USDA, 2018). For instance, in spite of having CBG as a common precursor, Δ9-THC, CBD, and CBC have dramatically different physiological effects. In the case of the two most widely studied compounds, Δ9-THC is known to produce euphoria and appetite stimulation (Volkow et al., 2014)**,** while CBD is non-euphorigenic and is thought to be anti-epileptic (Jones et al., 2010) and anti-inflammatory (Carrier et al., 2006)**.** Much less is known about CBG and CBC, but since there are differences in ring structure it is not surprising that they have differing pharmacological properties; however, cannabinoids other than Δ9-THC and CBD have had little exploration and characterization.

Interest in alternative cannabinoids

Recent events are prompting increased interest in the pharmacotherapeutic potential of the 100+ cannabinoid compounds. First, there exists growing belief that the beneficial effects of Cannabis derive from the entourage effect: all of the *Cannabis*-derived cannabinoids, terpenoids, and flavonoids acting in concert. While there is little evidence to document this, it does support the notion that the entire plant material needs to be consumed. Then, with the explosion in sales of CBD products, entrepreneurs are searching for the next economic market, and it appears that CBG oil may prove to be that market. Indeed, several commercial outlets are advertising CBG as the "mother of all cannabinoids" - presumably because in *Cannabis* it is a common precursor.

However, CBG will not give rise to the myriad other cannabinoids when taken into the human body because of the lack of necessary conversion pathways.

Work in recent years has garnered interest in other cannabinoids such as CBC and Δ9-THCV (tetrahydrocannabivarin), but more studies are needed. Δ9-THCV is suggested to induce similar effects as Δ9-THC without excessive appetite stimulation (Tudge et al., 2014), and studies of CBC show it can improve inflammation-related pain (Wirth et al., 1980).

In the present review, we explore potential therapeutic benefits and adverse side effects of CBG. Recent literature on CBG has revealed that its pharmacology addresses therapeutic targets distinct from those of Δ9-THC and CBD. We summarize previous investigations into therapeutic applications and propose new areas of interest for CBG's medical use. *In vitro* and *in vivo* studies suggest a potential future for CBG to address unmet needs in medical therapy, including its actions on adrenergic, serotonergic, peroxisome proliferator-activated receptor (PPAR), and cannabinoid receptor families (Cascio et al., 2010; Rock et al., 2011; O'Sullivan, 2016; Navarro et al., 2018).

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9. Discussion

The Unique Pharmacologic Properties of CBG

CBG is distinct from Δ9-THC and CBD in its pharmacological profile as summarized in Tables 1 and 2. In a variety of different ways, CBG seems to reside, pharmacologically, in between Δ9-THC and CBD. From the receptor binding data presented in Table 1, CBG is more like Δ9-THC at the CB1/CB2 receptors than CBD, but with a lower affinity (by a factor of between 5-fold and 27-fold) (Pertwee, 2008; Cascio et al., 2010; Pollastro et al., 2011; Rosenthaler et al., 2014; Navarro et al., 2018; Navarro et al., 2020). However, a 2012 study of CBG in human cell culture demonstrated negligible binding affinities for CBG at CB1 and CB2 receptors (Granja et al., 2012). More research is required to better understand the effects of CBG *in vivo* on cannabinoid receptor function and activity. In addition, CBD and CBG are very comparable at six transient receptor potential cation channels (TRPA1, TRPV1, TRPV2. TRPV3, TRPV4, and TRPM8) with relatively minor differences in affinity (generally less than 5-fold differences) (De Petrocellis et al., 2011; Pollastro et al., 2011; De Petrocellis et al., 2012; Muller et al., 2018). Important differences or gaps in our knowledge, however, exist for three key players. First, for GPR-55 (the potential non-homologous CB3 receptor) there is no information on CBG binding (Ryberg et al., 2007). Second, CBG appears to be a very potent (nM to sub-nM affinity) agonist at the alpha-2 adrenoceptor (Cascio et al., 2010). Physiologically, this is so potentially important, that it is the subject of extensive discussion below. At the present time, there are no data on this receptor for CBD and Δ9-THC. Finally, there is a clear differentiation of CBG and CBD at the 5-HT_{1A} receptor, where the former is reported to be an antagonist while the latter is an indirect agonist (with an unreported affinity) (Russo et al., 2005; Cascio et al., 2010; Rock et al., 2011; Rock et al., 2012). There are currently no data in this regard for Δ9-THC.

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Table 2 provides a more detailed analysis of the physiological activities that are exhibited by CBG (whether in intact animals, whole cells, or sub-cellular preparations). CBG appears to act as an agonist at the alpha-2 receptor (with varying EC_{50} values reported) and all of the TRP family channels (except TRPM8, at which it is reported to act as an antagonist). Together, these data suggest that the physiologic effects of CBG tend towards Gi-mediated inhibition, auto-regulatory activity, and calcium-based signaling both by ion channels and PKC. Given the differential pharmacological profiles, there are a number of important concerns to consider concerning the therapeutic potentials and potential adverse outcomes for the widespread and unregulated use of CBG.

The Alpha-2 Adrenergic Receptor (the α2-Adrenoceptor)

The observation that CBG is a potent agonist at the alpha-2 receptor has significant implications for potential therapeutic uses and adverse side effects (Cascio et al., 2010). Catecholamines (dopamine, norepinephrine and epinephrine), produced by the nervous system, use a variety of methods to exert different physiologic effects. The main receptor families that mediate these functions are the adrenergic receptors (alpha [α-1 and α-2] and beta [β-1, β-2, and β-3] families) and the dopamine receptors (D1 through D5 receptors) (Molinoff, 1984). Using the norepinephrine system as an exemplar, these receptors almost all act in the following way: vesicles containing norepinephrine are released from the presynaptic neuron and synaptic norepinephrine activates postsynaptic G-protein coupled receptors (GPCRs) to create a unique downstream effect. While the catecholamine receptor families are all GPCRs of varying functions, the alpha-2 receptor class is best known as a presynaptic receptor (Saunders and Limbird, 1999), although it also functions in peripheral tissues and cells. Extensive study of this

autoregulatory receptor has shown its ability to dampen sympathetic nervous system activity through its Gi (inhibitory) activity that is triggered when synaptic norepinephrine binds to the presynaptic receptor. Additionally, alpha-2 receptor activation demonstrates other mechanisms of reduced sympathetic activity such as opening receptor operated K+ channels and inhibiting voltage-gated calcium channels (Cotecchia et al., 1990; Saunders and Limbird, 1999). The family of alpha-2 receptors has its own further classification into 3 receptor subtypes, alpha-2A, alpha-2B, and alpha-2C, the genes for which are found on chromosomes 10, 2, and 4, respectively (Saunders and Limbird, 1999).

While highly related, the different subtypes of the alpha-2 receptor are distinct in their sequence, structure, and receptor distribution in the body (Saunders and Limbird, 1999). Structural differences between subtypes arise from the intra- and extracellular regions of the receptor structure, as well as differential post-translational modifications. Anatomical differences are well characterized for the 3 subtypes in the brain and in the periphery through in-situ hybridization of receptor mRNA and through immunochemical analysis of rodent receptor locations (Saunders and Limbird, 1999). In the brain, 2A receptors are widely distributed and abundant in the locus coeruleus and in brainstem regions with homeostatic functions. Alpha-2B receptors are found in the thalamic nuclei, while 2C is found in the basal ganglia, olfactory tubercle, hippocampus, and cerebral cortex (Molinoff, 1984; Arnsten et al., 1988; Saunders and Limbird, 1999). The 2A and 2C receptors are the primary contributors of alpha-2 receptor function in the CNS. While all 3 subtypes are found in the nervous system, each subtype has a unique peripheral tissue distribution. In the periphery, 2A is found in platelets, beta cells of the pancreas, adrenal glands, intestinal epithelia, vascular endothelium, and smooth muscle cells (Molinoff, 1984; Saunders and Limbird, 1999). Alpha-2B is found in rat neonatal lung and liver,

the adult kidney, vascular endothelium, and smooth muscle cells, while 2C is only found in the adult kidney (Molinoff, 1984; Saunders and Limbird, 1999).

Many of the discoveries involving alpha-2 receptor function were derived from experimental administration of known alpha-2 agonists, such as clonidine, and measuring local and systemic effects. Current therapeutic interest in peripheral alpha-2 agonism effects center around their anti-hypertensive, sedative, and analgesic functions. Clonidine and dexmedotomidine (a newer and more selective alpha-2 full agonist) are often used in anesthesia settings (Hunter et al., 1997; Gertler et al., 2001). Clonidine was originally popular as an anti-hypertensive agent but is now considered second-line or third-line in an anti-hypertensive regimen due to its effects at other non-adrenergic receptors. The vasodilatory and hypotensive effects of clonidine are multifactorial, owing to clonidine's actions at not only the alpha-2 receptors, but also imidazoline receptors, both of which reduce vascular tone when activated (Ernsberger et al., 1990). Another alpha-2 agonist, guanfacine, has higher selectivity at the alpha-2A receptor and less activity at imidazoline receptors. When compared to clonidine administration, guanfacine administration reduces blood pressure less than clonidine (Arnsten et al., 1988). While alpha-2 agonists are not indicated in most anti-hypertensive regimens today due to efficacy of other drugs, clonidine and guanfacine are commonly prescribed for neuropsychiatric diseases due to their effects on alpha-2 receptors in the prefrontal cortex. The role of alpha-2 agonists in neuropsychiatric disease depends on their ability to modulate and improve impaired prefrontal cortex (PFC) functioning (Arnsten et al., 1988; Arnsten, 2010). PFC impairment is a common finding during normal aging as well as conditions like attention-deficit hyperactivity disorder (ADHD), tic disorders, post-traumatic stress disorder, dementia, and others (Arnsten, 2010). Alpha-2 receptors, specifically 2A, are heavily involved in

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norepinephrine signaling in the PFC. Alpha-2 agonists are used to improve working memory and planning ability in ADHD in children and adults along with tic disorders and reduction of opiate withdrawal symptom severity (Arnsten, 2010). Alpha-2 agonists in the treatment of ADHD have the additional benefit of being effective alternatives or adjuncts to the first line treatment: stimulant medications. Use of alpha-2 agonists in conjunction with stimulant medications can reduce stimulant-induced tics and hypertension, along with reducing the necessary dosing to achieve symptom management for stimulant-sensitive patients (Arnsten et al., 1988; Arnsten, 2010). Clonidine and guanfacine, both alpha-2 agonists, have gained popularity in treatment of psychiatric disorders and are another example of alpha-2 subtype specificity determining functionality; guanfacine has less intense hypotensive and sedative side effects than clonidine while boasting increased activity at the PFC (Arnsten, 2010). Researchers believe that this is due to its specificity at alpha-2A. Interestingly, while agonism of alpha-2A receptors has shown benefits in selected psychiatric disorders, antagonism of alpha-2C receptors may be beneficial in other psychiatric disorders such as psychosis and schizophrenia (Uys et al., 2017). This diversity in receptor distribution and potential function indicates a major need for further research into subtype-specific actions and functions of the alpha-2 adrenergic receptors.

While alpha-2 agonists have considerable therapeutic applications, the current knowledge of CBG activity at alpha-2 receptor subtype and location is lacking. From the potency of CBG at this adrenergic receptor, ingestion may unpredictably change blood pressure, induce sedation, and interact with other cardiovascular medications. Later in this review we discuss the imperatives for further research into these physiologic effects of CBG administration.

The Serotonin 5-HT_{1A} Receptor

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Serotonin, or 5-hydroxytryptamine (5-HT), is a monoamine neurotransmitter produced throughout the body for a variety of physiological and neurological functions. It plays a central role in maintaining homeostatic functions (e.g., in the enteric gastrointestinal nervous system) (Coates et al., 2017). In the central nervous system, it is a target of anti-depression medications. Serotonin binds to different receptor families (Nichols and Nichols, 2008), and here we will focus on the 5-HT_{1A} receptor, which has previously been shown to interact with endogenous and exogenous cannabinoid ligands. The 5-HT_{1A} receptor is a Gi/o GPCR that inhibits adenylate cyclase, but the receptor has also been shown to interact with other growth factor pathways (Rojas and Fiedler, 2016). The receptor is located both pre- and post-synaptically, and the downstream functions of receptor activation vary based on neuronal identity and location. Inhibition of 5-HT_{1A} autoreceptor (presynaptic) activity is suggested to significantly affect the speed and efficacy of other serotonin-modulating drugs, such as selective serotonin reuptake inhibitors (SSRIs) (Artigas et al., 1996). A selective 5-HT_{1A} antagonist, WAY100635, has potentiated the effects of fluoxetine, an SSRI, *in vivo*. Additionally, the β-adrenergic/5-HT_{1A} antagonist pindolol was found to enhance the effects of several different classes of serotoninergic antidepressants in patients with major depressive disorder (Artigas et al., 1994). One possible mechanism for these effects is 5-HT_{1A} antagonists reduce the increase in 5-HT_{1A} autoreceptor activity created by antidepressants which increases synaptic serotonin availability.

In the present context, we note that CBG has been reported to be a potent (50 nM) 5-HT_{1A} antagonist. Given this pharmacological characteristic, it may have unpredictable potentiating effects on concurrently administered psychiatric medications and serotonin-modulating substances. Recently, Echeverry and colleagues reported that CBG and CBD have neuroprotective effects against oxidative neurotoxicity through a 5-HT_{1A} receptor mediated

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mechanism (Echeverry 2020). The effects of CBG on serotonin signaling or availability have not yet been thoroughly studied and more research is needed to understand the impact of CBG-mediated 5-HT_{1A} antagonism before it is made widely available in an unregulated commercial environment.

The Peroxisome Proliferator-Activated Receptors (PPAR)

The PPAR family is a collection of nuclear receptor transcription factors; there are three isoforms PPARα, PPARβ, and PPARγ. PPAR nuclear activation induces conformational changes and binding to PPAR response elements of DNA to modulate gene transcription (O'Sullivan and Kendall, 2010; O'Sullivan, 2016). These receptors transcriptionally regulate lipid metabolism, hepatic metabolic functions, and inflammation. PPARs accept a wide variety of ligands due to their large binding domains, and many cannabinoids and their metabolites are reported to interact with the various isoforms (O'Sullivan and Kendall, 2010; O'Sullivan, 2016). In the reviewed studies, CBG exhibits stronger affinity to the PPARγ receptor than Δ9-THC and CBD.

PPARγ will regulate adipocyte differentiation, insulin sensitivity, and inflammatory states. This is the therapeutic mechanism of thiazolidinediones, like rosiglitazone, commonly used in patients with type 2 diabetes to improve adipocyte functioning and increase insulin sensitivity (O'Sullivan and Kendall, 2010; O'Sullivan, 2016). Cannabinoid compounds display varying affinities at PPAR isoforms, and the effects of CBG on these receptors are being studied in the regulation of inflammation and metabolic functioning; these potential therapeutic effects are reviewed in the next section.

Potential Therapeutic Potentials for CBG

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Based on the receptor signaling of the alpha-2, 5-HT_{1A}, and PPARɣ receptors and the reported affinities of CBG at these receptors (in the tens of nM to sub-nM range), there are many reasons to believe that CBG will have therapeutic potential (Cascio et al., 2010; Rock et al., 2011; Granja et al., 2012). Similarly, however, there are reasons to monitor high dose CBG for untoward side effects beyond drug-drug interactions.

Neuroprotection and Neuromodulation

A number of studies have shown that CBG and a second generation synthetic quinone derivative, VCE-003.2, have neuroprotective potential *in vitro* and in animal models to reduce the severity of neurological illnesses such as Huntington's disease (HD), amyotrophic lateral sclerosis (ALS), Parkinson's disease (PD) and multiple sclerosis (MS); this seems to largely be mediated through PPARɣ (Granja et al., 2012; Carrillo-Salinas et al., 2014; Díaz-Alonso et al., 2016; Mammana et al., 2019). VCE-003.2 is distinct from the first generation CBG quinone derivative, VCE-003 because it retains neuroprotective activity through dose-dependent PPARɣ activation, but does so without the cytotoxicity and thiophilic properties of VCE-003 (Díaz-Alonso et al., 2016). These properties lend some pharmacological explanation to its beneficial effects on various models of neurotoxicity. Indeed, VCE-003.2 is reported to have negligible affinity at either CB1 or CB2 (>40 μM). CBG and VCE-003.2 have been shown to reduce the inflammatory molecules TNF-α, IL-1β, IL-6, MIP-1α, and PGE2 in rat microglial cells treated with LPS, and both compounds reduce glutamate-induced oxidative cell death in mouse hippocampal cells (Granja et al., 2012).

Using two mouse models of HD, Valdeolivas and colleagues reported improvement of HD symptoms (Valdeolivas et al., 2015). In particular, in 3-nitropropionate-treated mice, CBG prevented striatal neuron death, reduced markers of inflammation, and improved motor deficits

(Valdeolivas et al., 2015). The results in a transgenic R6/2 mouse model of HD were not as robust. Treatment with CBG did not reduce the weight loss observed in this model, but it did moderately improve rotarod performance and reduced the number of huntingtin aggregates (Valdeolivas et al., 2015). A study using VCE-003.2 found a similar improvement in HD symptoms in the murine model of HD (Díaz-Alonso et al., 2016). In the quinolinic acid murine model of HD, treatment with VCE-003.2 improved rotarod performance (Díaz-Alonso et al., 2016). VCE-003.2 also improved motor deficits in the NP3 model of HD, and this was found to be dependent upon PPARɣ activity in that inhibition of PPARɣ by T0070907 blocked the effect of VCE-003.2 treatment (Díaz-Alonso et al., 2016).

Experiments using the SOD^{G93A} mouse model of ALS found that VCE-003.2 improved the neuropathological symptoms, including: attenuating weight loss, improved clinical scores, and preservation of motor neurons in the spinal cord (Rodríguez-Cueto et al., 2018). Additionally, astrocytes cultured from SOD^{G93A}, when treated with VCE-003.2, exhibited a morphology close to wild-type astrocytes (Rodríguez-Cueto et al., 2018). Using an LPS-induced inflammatory model of PD, VCE-003.2 reduced the inflammatory markers TNF-α, IL-1β, and iNOS, and this response was mediated through PPARɣ (García et al., 2018). Finally, VCE-003.2 has also been shown to improve motor activity, reduce VCAM-1 expression, and decrease microglia activity, in the Theiler's murine encephalomyelitis virus model of MS (Granja et al., 2012).

Another area of interest is the therapeutic potential of CBG involving $5\text{-}HT_{1A}$ receptor activity. In 2020, Echeverry and colleagues studied the effects of CBG and CBD on neurotoxicity as assessed in neural cell cultures (Echeverry et al., 2020). The neuroprotective qualities of CBD and CBG were distinct at two different models of oxidative damage (the H_2O_2

model and the rotenone model) (Echeverry et al., 2020). The authors found that the protective effects were lost when CBG and CBD were administered with a 5-HT_{1A} antagonist, WAY-100635, but were unchanged with CB1 and CB2 receptor antagonists (Echeverry et al., 2020). This suggests that the protective effects of CBG and CBD against oxidative neurotoxicity are derived from a 5-HT_{1A} receptor mediated process (Echeverry et al., 2020).

Finally, a relatively unexplored area of CBG neuromodulation is its effect on neurotransmission. Recently, transcriptomic changes by administration of CBD and CBG on motor neuron-like cells has shown an insight into this neuromodulatory effect. In NSC-34 motor neuron like cells, authors Gugliandolo and colleagues compared the transcriptomic changes from CBD and CBG. The two compounds similarly changed transcription in the dopamine, GABA, and glutamate pathways (Gugliandolo et al., 2020). This was from, generally, reduced expression of glutamate release genes, enhanced expression of GABA release genes, and upregulated dopamine D4 receptor and its downstream effectors (Gugliandolo et al., 2020). Characterization of these changes on behavior and neuronal signaling through translational research is necessary to better understand the context of CBG in psychopharmacology. However, the results do speak to the need to monitor untoward side effects.

Gastrointestinal Disease

Cannabigerol has been explored as a therapeutic for gastrointestinal diseases such as colorectal cancer and colitis using mouse models. In the dinitrobenzene sulfuric acid (DNBS) model of colitis, treatment with CBG increased the rate of tissue recovery in the colon as measured by histological structure, the ratio of colon weight to length, colonic permeability, and reduced inflammation (Borrelli et al., 2013). The authors also found that CBG was also effective as a

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treatment to prevent colitis-associated damage (Borrelli et al., 2013). In a follow-up study, it was found that cannabigerol reduced tumor formation in the azoxymethane (AOM) model of colorectal cancer and reduced xenograft tumor growth (Borrelli et al., 2014). While the authors conclude that this reduction was mediated by TRPM8, these studies were performed in cell culture, rather than in the murine model. In 2020, Pagano and colleagues used a mouse model of DNBS-induced colitis to study the effects of CBG on inflammatory activity. Orally administered CBG reduced colonic inflammation as measured by significantly reducing MPO activity, IL-1b levels, and serum FITC-dextran concentration (Pagano et al., 2020). In contrast, CBD on its own did not induce significant changes to these metrics. Adding fish oil to the treatment paradigm increased the CBG-induced reduction in MPO activity, IL-1b levels, and serum FITC-dextran concentration (Pagano et al., 2020).

CBG has also been shown to increase feeding in rats and to reduce weight loss associated with cisplatin chemotherapy (Brierley et al., 2016; Brierley et al., 2017; Brierley et al., 2019). However, an earlier study found no impact of CBG on feeding behavior (Farrimond et al., 2012). Unlike Δ9-THC and CBD, CBG has not been found to have antiemetic effects and appears to oppose the antiemetic effects of CBD (Rock et al., 2011). Taken together, these studies indicate that there may be a role for cannabigerol in chemotherapy-associated weight loss and loss of appetite, although Δ9-THC (Dronabinol) and Nabilone have already been proven efficacious, CBG lacks the potential euphoric side effect of these medications.

Metabolic Syndrome

Metabolic syndrome affects millions of Americans and contributes to the highest burden of healthcare costs and preventable mortality in the country. A clinical diagnosis, metabolic

syndrome is a combination of insulin resistance, obesity, hypertension, high levels of low-density lipoprotein (LDL) and reduced levels of high-density lipoprotein (HDL). While a wide array of medications and surgical interventions are available for metabolic syndrome, few treatments are effective enough to serve as monotherapy, and many patients need multiple medications with harsh side effects to keep this chronic illness at bay.

Recent studies on CBG provide promise for its use as part of a multifactorial pharmacotherapy for metabolic syndrome and its components. Hypertension, one component of metabolic syndrome, can be modestly reduced with alpha-2 agonist therapy, which reduces synaptic norepinephrine levels to reduce vasoconstriction and improve blood pressure. CBG is currently the only known cannabinoid that is an agonist at the adrenergic receptor (Cascio et al., 2010). In addition, as previously mentioned, CBG and its derivatives are shown to act on PPARγ receptors in its role on neuroinflammation. In 2019, *in vitro* modeling of phytocannabinoids suggested CBG as a dual PPARα/γ agonist first through computer modeling and prediction, and then confirmation in HepG2 (human liver epithelial-like) and 3T3L1 (mouse embryo fibroblast) cell lines (D'Aniello et al., 2019).

More support for PPARα/γ agonism of CBG came in 2020 from researchers who tested PPARα/γ-dependent differentiation in bone marrow mesenchymal stem cell lines (Fellous et al., 2020). This effect was enhanced when combined with CBD, and had a similar effect as a commonly prescribed drug for metabolic syndrome, rosiglitazone (Fellous et al., 2020). Rosiglitazone is known to act as an agonist at PPARy receptors and to increase expression of GLUT4 glucose channels; both of these effects increase insulin sensitivity. CBG and the CBG/CBD combinations provided similar effects *in vivo* as rosiglitazone, and additive effects to improve adipogenesis (Fellous et al., 2020). Renewed adipogenesis and insulin sensitization

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both improve symptoms and pathology of metabolic syndrome; therefore, CBG could be explored as a potential therapeutic for this devastating disease.

Antibacterial Agent

A number of cannabinoids have been reported to have antibacterial activity; however, CBG was found to be among the most potent cannabinoids tested against antibiotic resistant strains of Staphylococcus aureus (Appendino et al., 2008). In comparison to conventional antibiotics, CBG had a lower minimum inhibitory concentration than norfloxacin in 5 of the 6 strains tested, and was more potent than erythromycin, tetracycline, and oxacillin in at least one resistant strain (Appendino et al., 2008). Using a systemic S. aureus infection model in mice, Farha and co-workers showed that CBG was as effective at reducing colony forming units as vancomycin (Farha et al., 2020). This study also found that CBG was effective against gram-negative bacteria only when the outer membrane was first permeabilized, suggesting that the outer membrane of gram-negative bacteria presents a permeability problem for cannabinoids (Farha et al., 2020). Using *in silico* modeling, Pinzi and colleagues found that CBG acts as an inhibitor of enoyl acyl carrier protein reductase (InhA), and they verified their model with *in vitro* testing and found CBG inhibits enoyl acyl carrier with an IC_{50} value in the low micromolar range (Pinzi et al., 2019). These data are encouraging because there is a need to develop novel therapeutics as antibiotic resistance in bacteria is a continuing healthcare issue.

Potential Arguments Against CBG as a Therapeutic

We have highlighted the potential of cannabigerol as a therapeutic and in medical research; however, for it to be seriously considered as a potential therapeutic, it must be rigorously tested for safety and unintended effects.

CBG has potent activity at the alpha-2 receptor and this unique property could also induce unintended cardiovascular consequences like hypotension, bradycardia, and xerostomia. Additionally, some investigators have reported hypertension as a counterintuitive adverse effect in high doses of alpha-2 agonists, which appears to be mediated by the alpha-2B receptor subtype (Philipp et al., 2002). The potential for this adverse effect is unclear in the case of CBG since its activity at different alpha-2 receptor subtypes has yet to be studied. While we surmise that CBG may have therapeutic potential among neurological, gastrointestinal, and metabolic disorders, there must be more research to ensure that unintended cardiovascular effects do not reduce the utility of CBG. In addition, in this era of unregulated CBD preparations, companies are making unsubstantiated claims and over-selling the benefits and under-selling the risks. Indeed, companies are already touting CBG as the "mother of all cannabinoids" presumably because it is the immediate precursor of CBD and Δ9-THC. What they fail to point out, however, is that only the *Cannabis* plant goes on to convert CBG to the other molecules – the human body does not. This is problematic because very few *Cannabis* strains actually harbor large concentrations of CBG, so there is not much prior reporting (and virtually no documentation) of human side-effects of CBG.

Several recent cautionary situations come to mind. In a couple of recent reports on CBD oil, it has been reported that not all CBD oil preparations had concentrations of CBD close to what the manufacturer claims (Bonn-Miller et al., 2017; Pavlovic et al., 2018; Raup-Konsavage et al., 2020; Urasaki et al., 2020). Moreover, the US Food and Drug Administration had to

prompt recalls for CBD preparations that contained unacceptable concentrations of lead (FDA, 2020a; FDA, 2020b). We have reported that, even when the different CBD oil preparations contain the reported levels of this phytocannabinoid, they had very different activities in suppressing cancer cell growth *in vitro* and only one approximated the activity of pure CBD (Raup-Konsavage et al., 2020). Finally, there is also the very real risk of inducing drug-drug interactions when over-the-counter preparations like CBD oil or CBG oil are taken by a patient on other (prescription) medications (Kocis and Vrana, 2020). This may lead to unintended adverse consequences when not appropriately monitored.

Future Imperatives

Research into cannabigerol is in its infancy but has shown promise for addressing a diverse array of therapeutic needs. Based on its pharmacodynamics, here we highlight potential indications for CBG and its derivatives to improve available drug treatment regimens for selected diseases and medical conditions. However, these applications will rely on additional research studies to further understand how CBG can be used safely and effectively.

First, there is potential for CBG as a major player in the treatment of metabolic disease as described by its action on the PPAR family of receptors to improve insulin sensitivity and adipogenesis. A supplementary effect is its anti-hypertensive properties at alpha-2 receptors. Diabetic patients frequently have hypertension, hyperlipidemia, and insulin resistance stemming from glucose dysregulation and vascular endothelial dysfunction. CBG may improve this profile as an adjunct to the mainstay of treatment, metformin, or potentially serve as its own regimen. Alpha-2 agonists such as clonidine are infrequently used as anti-hypertensive agents in current practice, largely because these effects of clonidine are enhanced with imidazoline receptor

activity; this rendered clonidine too powerful as an anti-hypertensive drug and unreliable in practice. To our knowledge, the sympatholytic effect of CBG is limited to alpha-2 activity, which makes it more useful for this indication than clonidine, but more studies are needed.

Second, several studies have described the neuroprotective effects of CBG through action on the PPAR family of receptors. Other sources have reported reduction in age-related cognitive decline in neurodegenerative disease patients with the addition of alpha-2 agonists to their treatment regimens. While CBG's effect on cognition has yet to be studied, it may play a role for improving quality of life in these vulnerable populations, as the few drugs currently available for neurodegenerative diseases also carry uncomfortable and disabling side effects.

Third, similar to other phytocannabinoid derivatives, CBG may play an important role for improving the drug cocktails of patients who struggle with disorders of executive function, such as schizophrenia and ADHD. Two current alpha-2 agonists, clonidine and guanfacine, are indicated for their alpha-2 mediated action on the human prefrontal cortex to improve executive function and self-regulation; however, clonidine is less safely prescribed due to its potent anti-hypertensive properties and generalized action on alpha-2 receptor subtypes. Guanfacine is well-studied as an adjunct therapy with stimulants, in ADHD, due to its alpha-2A receptor subtype specificity. The subtype specificity of CBG has yet to be elucidated; therefore, it cannot be predicted how CBG will improve executive dysfunction compared to guanfacine. Finally, researchers have studied the effects of CBG as a safe appetite stimulant in chemotherapy-related appetite suppression *in vivo* and as an agent that reduces *in vitro* signs of pathology in colitis and colorectal cancer.

In closing, while there is much to suggest that CBG may provide alternative therapeutics for a number of disorders, much is left to learn. In particular, given the potent bioactivity CBG

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displays in a number of settings, we should be very cautious about releasing it in an unregulated retail environment. There is simply insufficient experience with this relatively rare phytocannabinoid and the potential for adverse effects is high. Given the dramatic increase in unregulated CBD oil use following deregulation of hemp, it behooves the pharmacology community to undertake CBG research before its use explodes as well.

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11. Authorship Contributions

Designed and wrote the manuscript: Nachnani, Raup-Konsavage and Vrana

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13. Footnotes

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14. Figure and Table Legends

Figure 1: Biosynthesis Pathway of the Primary Cannabinoids

Cannabinoid biosynthesis begins with the combination of geranyl pyrophosphate and olivetolic acid to form cannabigerolic acid (CBGA). CBGA serves as the substrate for the synthesis of tetrahydrocannabinolic acid (Δ9-THCA) and cannabidiolic acid (CBDA). Decarboxylation of CBGA, Δ9-THCA, and CBDA by heat results in cannabigerol (CBG), tetrahydrocannabinol (Δ9-THC), and cannabidiol (CBD), respectively. Because CBGA serves as the substrate for the synthesis of the major cannabinoids very little is typically found in material from *Cannabis sp*.

Table 1: Pharmacodynamic Properties of Δ9-THC, CBD, and CBG at Cannabinoid Receptors

Binding affinities for Δ9-THC, CBD, and CBG at the two canonical cannabinoid receptors (CB1 and CB2) as well as a third receptor, GPR55 (commonly referred to as CB3). Δ9-THC acts as an agonist at all three receptors while CBD acts as an antagonist, CBG acts as a weak or partial agonist at CB1 and CB2, while its function at GPR55 is currently unknown ([1]Ryberg et al., 2007; [2]Pertwee, 2008; [3]Cascio et al., 2010; [4]Pollastro et al., 2011; [5]Rosenthaler et al., 2014; [6]Navarro et al., 2018). N.T., not tested.

Table 2: Pharmacodynamic Properties of Δ9-THC, CBD, and CBG at Non-Cannabinoid Receptors

Binding affinities for Δ9-THC, CBD, and CBG at TRP ion channels, alpha-2 adrenoceptors, the serotonin receptor 5-HT_{1A}, and PPARγ are presented. Values are all EC50 for agonists and IC50 for antagonists ([1]Russo et al., 2005; [2]Cascio et al., 2010; [3]De Petrocellis et al., 2011; [4]Pollastro et

al., 2011; [5]Rock et al., 2011; [6]De Petrocellis et al., 2012; [7]Granja et al., 2012; [8]Rock et al., 2012; [9]Muller et al., 2018). N.T. indicates “not tested” whereas N.D. indicates “not detected”.

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15. Tables

	Δ9-THC		CBD		CBG	
Receptor	**Affinity (nM)**	**Function**	**Affinity (nM)**	**Function**	**Affinity (nM)**	**Function**
CB1	5.1-80.3 (Ki)[2,5]	Partial Agonist	1458.5-4900 (Ki)[2,5]	Inverse Agonist/ Antagonist	440-1045 (Ki)[3,4,5,6]	Weak Agonist
CB2	3.1-75.3 (Ki)[2,5]	Agonist	372.4-4200 (Ki)[2,5]	Inverse Agonist	153.4-1225 (Ki)[3,4,5,6]	Partial Agonist
GPR55	8 (EC50)[1]	Agonist	445 (IC50)[1]	Antagonist	N.T.	Unknown

Table 1

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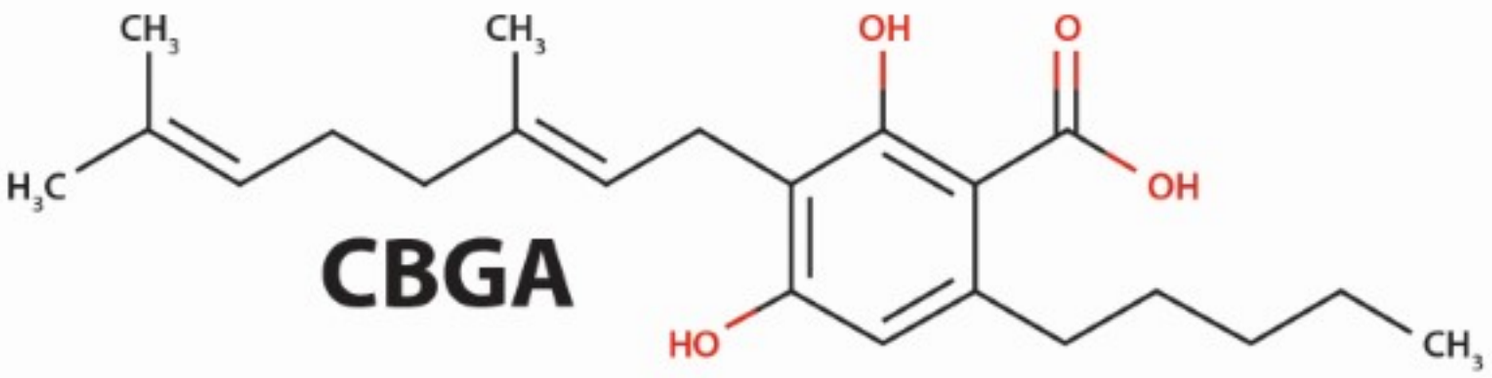
	Δ9-THC		**CBD**		**CBG**	
Receptor	**Affinity (nM)**	**Function**	**Affinity (nM)**	**Function**	**Affinity (nM)**	**Function**
TRPA1	230 [3,9]	Agonist	110 [3,9]	Agonist	700 [3,4,9]	Agonist
TRPV1	N.D. [3,9]	Unknown	1000 [3,9]	Agonist	1300 [3,4,9]	Agonist
TRPV2	650 [3,9]	Agonist	1250 [3,9]	Agonist	1720 [3,4,9]	Agonist
TRPV3	9500 [5,9]	Agonist	3700 [5,9]	Agonist	1000 [5,9]	Agonist
TRPV4	850 [5,9]	Agonist	800 [5,9]	Agonist	5100 [5,9]	Agonist
TRPM8	160 [3,9]	Antagonist	140 [3,9]	Antagonist	160 [3,4,9]	Antagonist
α2-adrenoceptor	N.T.	Unknown	N.T.	Unknown	0.2-72.8 [2]	Agonist
5-HT_{1A}	N.T.	Unknown	N.D. [1,8]	Indirect Agonist	51.9 [2,5]	Antagonist
PPARγ	2120 [7]	Agonist	2010 [7]	Agonist	1270 [7]	Agonist

Table 2

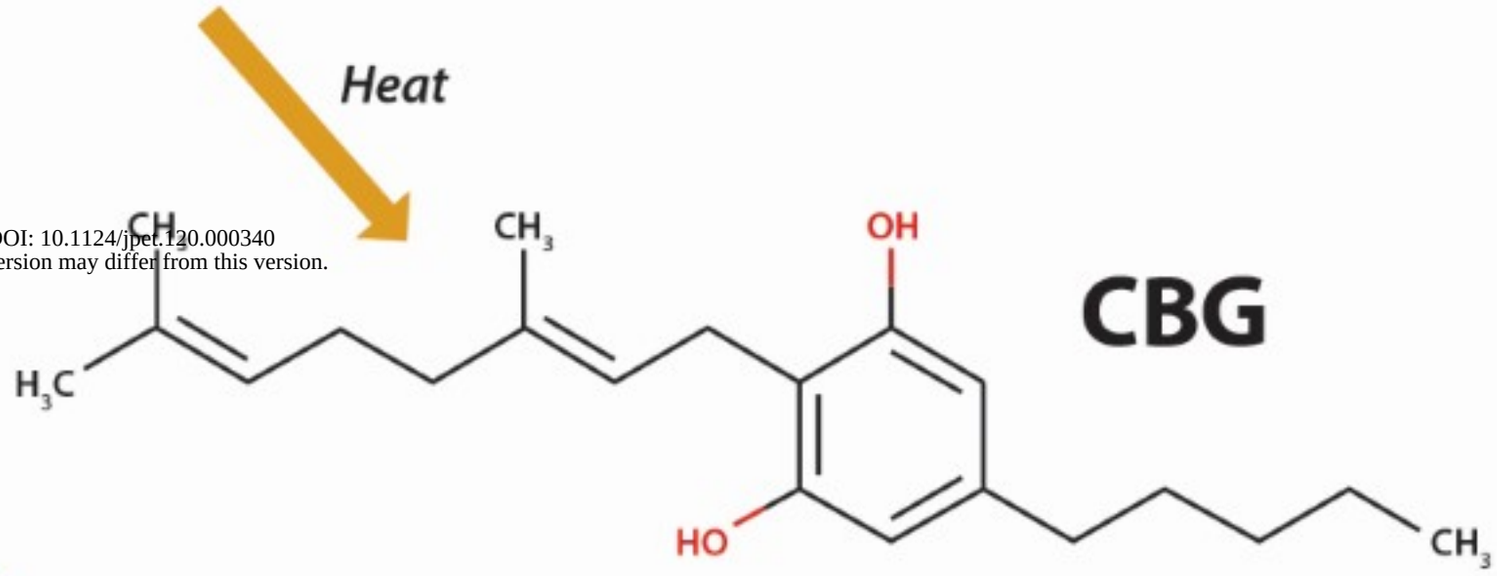
Geranyl Pyrophosphate **Olivetolic Acid**

Geranyl-pyrophosphate—olivetolic acid Geranyltransferase



CBGA

Heat



CBG

THCA Synthase

CBDA Synthase

Physiological/Psychological Activities:

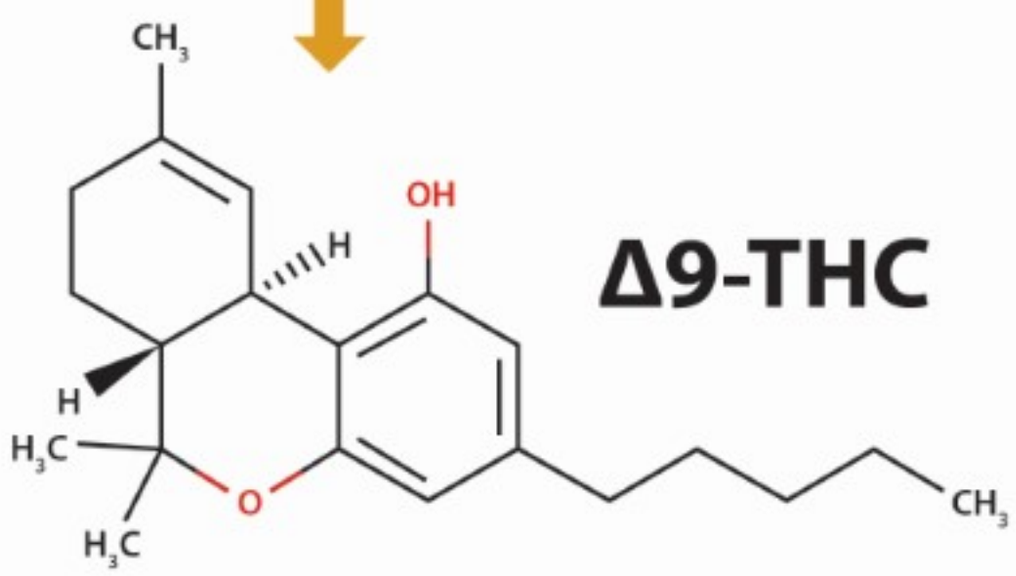
- Anti-Inflammatory
- Antibacterial
- Hypotension/Vasoconstriction
- Neuroprotective

THCA

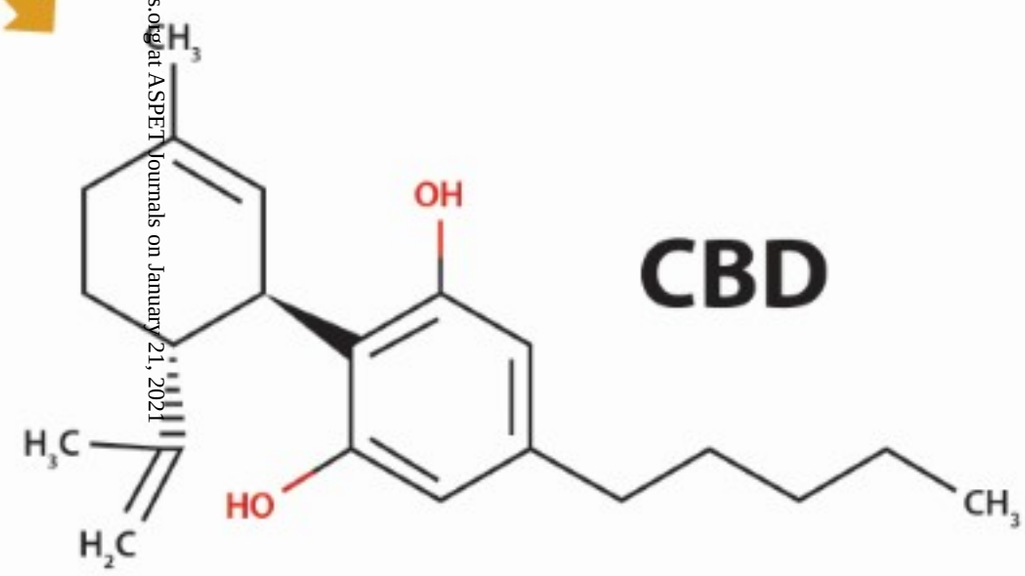
CBDA

Heat

Heat



Δ9-THC



CBD

Physiological/Psychological Activities:

- Euphorigenic
- Antiemetic
- Appetite Stimulant
- Analgesic

Physiological/Psychological Activities:

- Anti-Inflammatory
- Anticonvulsant
- Analgesic