The current state and potential direction of cannabis research

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ABSTRACT

* Cannabis sativa has long been linked as a medicinal treatment for many conditions such as cancer, epilepsy, bone fracture healing, anti-inflammatory response, neuro-degenerative disorders such as multiple sclerosis (MS), and rheumatoid arthritis. This article discusses the underlying conditions that are significantly controlled or modulated by cannabis. The primary components that have been isolated from cannabis are the psychoactive cannabinoid Δ(9)-tetrahydrocannabinol (THC) and cannabidiol (CBD). Both cannabinoids bind directly to endocannabinoid receptors that are located abundantly in the nervous system and visceral organs. Cannabis has also been used as a treatment to manage pain for chemotherapy patients and induce hunger in HIV (Human Immunodeficiency Virus) positive patients. Cannabis has received vast amounts of attention because of its effect on treating epilepsy. Although the biochemical pathway for this is not yet known, the current evidence in favor of this treatment has opened the door for further research. Finally, the cannabis terpenoids that have been isolated from Cannabis sativa are discussed as a future focal point for cannabis research. This review article sets to provide the medical community with a broad synopsis of recent research trials on Cannabis sativa that have indicated its medicinal benefits. The removal of the Public Health Service (PHS) Review, which has inhibited government funding and access to research samples, is an imperative step for obtaining further knowledge into the biochemical pathways and effects of medical cannabis.

KEY WORDS: Cannabis; Endocannabinoid; Cannabidiol; Tetrahydrocannabinol; Anti-Inflammatory Agent; Lysyl Hydroxylase

INTRODUCTION

Over the last twenty years, there has been a considerable amount of research on the impact of the molecular components of cannabis on human health and disease [1]. There has been a great amount of interest surrounding the legalization of cannabis all over the world. As federal, state and local governments look to create a compliant and transparent cannabis distribution and sales system, researchers are finding more funding opportunities to further study the bio-molecular components of cannabis and its medicinal benefits, such as its use in pain management for cancer patients undergoing chemotherapy [2]. Up until recent, scientists found it difficult to attain federal funding for cannabis research. In the United States, cannabis is federally illegal and remains on the list of Schedule II drugs. As a result, federal research funding has been scarce and virtually non-existent. But on June 22, 2015, the Obama administration removed the PHS (Public Health Service) Review and took steps to allow more researchers to attain federal funding and higher stockpiles of cannabis for research. In the past, the PHS Review significantly slowed the progression of cannabis research with strict requirements for the distribution of cannabis to research facilities. The PHS Review was originally set in place by the Clinton Administration to promote cannabis research, but the restrictions created large barriers that made it impossible for private research groups to gain access to cannabis or grow their own samples.

Despite the limitations that have been placed on cannabis research, remarkable advancements have been made to understand the bio-molecular components of cannabis and their metabolic effects on the human body. Within the last fifty years, the active cannabinoids of cannabis (THC and CBD) have been isolated and their effects on the human body’s endocannabinoid system (ECS) have been noted. Between 1963 and 1964, Dr. Raphael Mechoulam established the structure of CBD and discovered the psychoactive component of Cannabis sativa, THC [1]. Both THC and CBD are cannabinoids that activate or inhibit the endocannabinoid receptors. Thirty years after the discovery of THC and CBD, it was discovered that the human body developed its own endocannabinoids that contained a similar structure and mechanism of action as THC. These endogenous endocannabinoids are known as N-arachidonoylthanolamine (anandamide) and 2-arachidonoylglycerol (2-AG). This discovery opened the door for additional research on the body’s endogenous ECS, highlighting the significance of endocannabinoids on human physiology, health and disease.
The research on endogenous endocannabinoids also helped to identify a collection of proteins that bind, synthesize and degrade endocannabinoids. This organization of proteins, receptors and endocannabinoids comprise the ECS. Endogenous cannabinoids have been linked to many biological functions such as cell activation/deactivation, cell survival and death, and proliferation and differentiation of progenitor stem cells [1]. By stimulating the same system of internal receptors, exogenous cannabinoids from cannabis are believed to provide the same physiological effects on the body as the endogenous cannabinoids. Therefore, THC and CBD cannabinoids of the cannabis plant have been found to activate the ECS of the human body in a similar manner as the endogenous cannabinoids. Endocannabinoids have been found to impact several characteristics of the human body's pathophysiology. They are components with a favorable profile for drug safety [2]. Many scientists consider the discovery of cannabinoids to be of unprecedented importance, possibly the most dynamic signaling molecules ever found.

The following research paper discusses the current focus of cannabis research and the pathological conditions that are believed to be moderated through activation or inhibition of the ECS. The ECS uses G protein-coupled cannabinoid receptors (CB-1 and CB-2) for the activation or inhibition of a particular cell. The endocannabinoids target one or both of these receptors either as CB-1 receptor inhibitors or CB-2 receptor activators [3]. Cannabinoid receptors have been largely found throughout the body, CB-1 receptors among the nervous system, and CB-2 receptors among the body's peripheral organ cells. Exogenous cannabinoids that are found in cannabis, THC and CBD, lock into this network of existing receptors and provide the same metabolic functions as endogenous endocannabinoids anandamide and 2-AG.

ANTI-HYPERALGESIA, ANTI-INFLAMMATORY AND RHEUMATOID ARTHRITIS

Cannabis has been linked as a moderate pain reducer and anti-inflammatory agent, acting in a way that's similar to the endogenous cannabinoids (anandamide) on the ECS. The process of neuro-inflammation encompasses multifaceted biological routes that remove harmful stimuli and start the healing process of the damaged tissue [4]. The function of endogenous cannabinoids have been noted in multiple types of inflammation processes. More specifically, by interacting with CB-1 receptors, cannabinoids have shown to improve certain types of edema including carrageenan-induced edema, and anti-hyperalgesia effects such as carrageenan-induced thermal hyperalgesia. The results showed that the same anti-hyperalgesia effects were produced from the endogenous cannabinoid anandamide when 0.01 ng was administered to CB-1 receptors. Peripheral administration of anandamide showed to decrease hyperalgesia by interacting with a CB-1 cannabinoid receptors antagonist. A separate experiment demonstrated anandamide’s ability to inhibit edema by inhibiting CB-1 receptor activity [5].

CBD, which does not cause any psychoactive effects, has shown properties as an anti-inflammatory agent. Researchers have focused primarily on CB-2 receptors as a target to reduce the inflammation response. More specifically, the binding of CBD to CB-2 receptors in synovial joints has shown to reduce the synthesis of TNF-α, a pro-inflammatory cytokine [6]. The body’s anti-inflammatory response to cannabis has also been linked as a treatment for rheumatoid arthritis. Synovial tissues from rheumatoid joints contain a high number of CB-2 receptors, more than the tissue of osteoarthritis joints. A western blot analysis confirms that CB-2 agonists, such as CBD, inhibit the production of the anti-inflammatory agents such as interleukin-6 (IL-6), chemokine ligand-2 (CCL-2), and metalloproteinase-3 (MMP-3) from its biochemical predecessor (fibroblast-like synoviocytes) which are stimulated by the cell signaling protein TNF-α. As a result, administration of CB-2 agonists reduces the inflammation process by inhibiting cell infiltration, destruction of bone, and murine collagen type II IgG1 antibody production [7].

NEURO-DEGENERATIVE DISORDERS

Cannabinoids have also been the focus in research studies pertaining to neuro-degenerative processes such as MS and autoimmune encephalomyelitis [8, 9, 10]. The ECS affects both components of the nervous system, central and peripheral pathways [11]. In the brain, the signaling effect of the cannabinoid/receptor complex is primarily inhibitory. This suggests that cannabinoids play a vital role in combatting many types of CNS (Central Nervous System) diseases by inhibiting the release of certain neurotransmitters [12]. Recent studies have indicated that cannabinoids impede the advancement of neurodegeneration [13]. Because neuro-inflammation plays a significant role in neurodegeneration, CB-2 receptors have been targeted for the development of neuroprotective therapies [14]. Other neuropathies that cannabinoids have been linked to as a method of treatment include MS, spastic paralysis, pain, glaucoma, nausea and vomiting. Additionally research trials suggest that cannabinoids may also be used in the treatment of basal ganglia disorders such as dystonia, tics, tremors and epilepsy [15].

MS is often caused by viral or other environmental stimuli attacks on an individual with a prone genotype. The disease is considered to be an autoimmune condition that causes demyelination of the CNS. This process severely damages neurons and oligodendrocytes, the primary cells of the CNS that form myelin around neuronal axons. As a result, the nervous system becomes a micro-environment full of demyelinated axons and neuro-inflammatory effects. Cannabis has been widely used to treat the underlying symptoms of MS [16, 1]. Research trials have shown to inhibit signs of MS by modulating the activity of receptor-mediated (CB-1) neuron transmission [17, 18]. Other studies have indicated that cannabis is an effective treatment for slowing progressive neuro-degeneration [19].
THC, a partial CB-1 receptor agonist, induces immune-suppression [20]. More specifically, the CB-1 receptor mediated effect occurs secondary to activation on nerve cells instead of directly targeting immune cells [21]. These studies demonstrate that activation of cannabinoid receptors leads to the production of immune-suppressive molecules. Exogenous cannabinoids stimulate pre-synaptic CB-1 receptors that are found within the hypothalamus or other areas of the CNS, producing a downstream release of immune-regulatory molecules such as cortisol and sex hormones. These immune-regulatory molecules proceed to enter the bloodstream and reduce antigen presentation, pro-inflammatory cytokine production, and T-cells extravasation. This leads to a weakened peripheral auto-immune response and a reduction in inflammation [4].

BONE FRACTURE HEALING AND ENDOCHONDRAL SKELETAL GROWTH

The skeletal sympathetic nerve terminals on bone contain a high amount of CB-1 receptors which suggests that the ECS regulates bone formation [22]. Bone fractures are a common occurrence, leading an individual to experience an extended period of immobilization and discomfort. A study from 2015 reported that CBD enhanced the biomechanical healing properties in rat mid-femoral fractures [23]. The study demonstrated that maximal load and work-to-failure of rat femurs treated with a combination of CBD and THC for an 8 week period improved bone healing noticeably. The above-mentioned research trial consisted of three separate experiments. In the first experiment, rats with fractured femurs were either injected with THC or CBD to test the mechanical and structural properties of bone fracture healing. In the second experiment, the rats were injected with equal mixture amounts of THC and CBD to analyze the effects on a fracture callus. The first and second experiments monitored changes in molecular bone composition for eight weeks with the use of Fourier Transform Infrared Spectroscopy (FTIR). In the third experiment, the expression of THC and CBD on osteoblast enzyme lyl hydroxylase was analyzed [23]. Lysyl hydroxylase (oxygenase enzyme) is responsible for catalyzing the hydroxylation of lysine to hydroxylysine which induces collagen cross-linkage and stabilization in bone healing.

The first experiment concluded that stimulation of endocannabinoid receptors (CB-1) on osteoblasts with cannabinoids (CBD) improved the rate of bone healing. CBD markedly enhanced the mechanical properties and strength of the fractured bone. The second experiment, which tested the effects of THC and CBD administered simultaneously, concluded that THC increased the maximal force and stiffness of the bone slightly more than CBD. In the third experiment, CBD selectively increased levels of mRNA of the lysyl hydroxylase gene which led to faster rate of collagen cross-linkage and stabilization. Overall, this data showed that CBD improved fracture healing and demonstrated the mechanical role of collagen crosslinking enzymes in bone fractures [23].

Another study that was conducted on endocannabinoid receptors tested the ECS’s role in skeletal elongation. The study was performed as a result of the reduced height that is often observed in babies born from mothers who smoked cannabis during pregnancy. Researchers identified both of the endocannabinoid receptors, CB-1 and CB-2, expressed on hypertrophic chondrocytes of the epiphysial growth cartilage (ECG), which is involved in vertebral growth. More specifically, THC showed to slow skeletal elongation by inhibiting the role of CB-1 receptors in ECG, a role that’s primarily observed in femoral and lumbar elongation, resulting in reduced body weight. THC also showed to inhibit ECG chondrocyte hypertrophy and reduce hypertrophic cell zone thickness by influencing CB-2 receptors. Therefore, the study determined that there exists a local growth restraining the ECS system within ECG. Diacylglycerol lipase, and essential biosynthetic enzyme of the main ECS, and 2-AG were also found in abundance on hypertrophic chondrocyte cells of ECG [24]. Additional research is needed to determine the relevance of the present findings.

EPILEPSY

CBD is an anticonvulsant [25]. Epilepsy has received widespread recognition for its responsiveness to medical cannabis. Despite the mechanism of action not being fully understood, multiple cases have emerged of young children who suffer from epileptic seizures that reduce dramatically after treatment with CBD. Because THC and CBD act upon the cannabinoid receptors, sufficient data demonstrates the ECS’s role in generating, maintaining and controlling seizures [26]. There has also been a strong focus towards the treatment of pediatric epilepsy with CBD. A large amount of interest in CBD for the treatment of refractory epileptic attacks has yielded additional research trials in states where medical cannabis is legal [27]. A combination of anecdotal reports and limited lab trials has marked CBD as a strong treatment option for pediatric epilepsy. A high amount of attention has been focused specifically on Dravet syndrome, but there remains a lack of evidence on the effectiveness of CBD treatment for infantile spasms (IS) and Lennox-Gastaut syndrome (LGS) [28].

Although the mechanism of action for CBD treatment of epileptic seizures has yet to be isolated, substantial evidence has been gathered to suggest it as an effective medicinal option for refractory epilepsy [29]. But further research is needed to understand more about these mechanisms in order to develop other potential treatment options [30]. In a recent study of 117 parents with epileptic children (including 53 with IS or LGS), 85% of all parents reported reduced seizure frequency and 14% reported complete eradication of epileptic attacks after CBD treatment was administered. Perceived efficacy and tolerability were similar across all groups that were observed. In the study, CBD treatment was administered for 6.8 months at a dosage of 4.3 mg/kg/day. Although the study appears to
show viable evidence that CBD treatment significantly reduces the onset of epileptic seizures, more research is required to determine efficacy and safety [28].

CANCER, HIV, AND AIDS

The medicinal properties of cannabis have led to the development of cannabinoid medications such as nabilone, dronabinol and nabiximol. Dronabinol is used for the treatment of cancer patients who experience nausea due to chemotherapy and acquired immune deficiency (AIDS) patients who experience weight loss from anorexia. The US Food and Drug Administration has approved dronabinol as a treatment option for those conditions. Nabilone has been approved by the FDA as a treatment for nausea secondary to cancer chemotherapy. Nabiximol is commonly used to treat spasatic attacks due to MS and the debilitating pain associated with cancer [31]. The cannabinoids that are found in Cannabis sativa have been linked to treating the common conditions (nausea, pain, anorexia, weight loss) that are associated with cancer and AIDS.

Cancer patients are normally given methotrexate for chemotherapy. Methotrexate serves as a competitive inhibitor for enzymes that produce folic acid. As a result, methotrexate inhibits the synthesis of DNA, RNA, thymidylates and proteins. A common side effect that chemotherapy patients encounter is nausea. In a study that tested the effects of THC on nausea from chemotherapy treatment, 15 patients were given a high-dose of methotrexate with oral THC and smoked cannabis. The THC and smoked cannabis effectively reduced nausea and vomiting in 14 of the 15 patients compared with a placebo [31].

Medicinal cannabis has been used for the treatment of debilitating pain [33]. In one particular study, pain was induced through an intradermal injection of capsaicin. A moderate dosage of cannabis decreased pain, whereas a high dosage of cannabis increased pain. There was also a study in which vaporized cannabis was given to a patient who was taking extended-release opiates for chronic pain [31]. The patients’ pain was reduced after vaporized cannabis was administered. However, plasma opioid levels did not change.

Cannabinoids have also demonstrated to be beneficial in the treatment of HIV related peripheral neuropathy [3]. HIV positive patients commonly report using medical cannabis to induce appetite and reduce nausea due to medication. In a placebo-controlled clinical trial of HIV-positive individuals who smoked cannabis, the results demonstrated an increase in food intake and weight gain [31]. It’s been suggested that these affects are caused by an increase in the hormones leptin and ghrelin, and a decrease in the levels of peptide tyrosine which helps to regulate appetite.

FUTURE FOCUS ON CANNABIS TERPENOIDS

Phyto-cannabinoids and terpenoids are components of cannabis that are synthesized within the glandular trichomes of secretory cells [33]. There has been a significant amount of focus on the bio-molecular composition and reactions that are associated with cannabis terpenoids. For example, Leonitus leonurus is a member of the Lamiaceae/Labiatae Family, also known as the Mint Family. Leonitus leonurus has been widely used as an herbal medicine, both topically and orally, to treat many conditions such as hemorrhoids, eczema, skin rashes, boils, itching, muscular cramps, headache, epilepsy, chest infections, constipation, spider bites and snake bites. Of the 50 compounds that have been isolated from leonitus leonurus, mainly terpenoids, labdane diterpenes have been linked to the therapeutic effects previously mentioned [18]. Although limited evidence has been gathered in regards to Cannabis sativa terpenoids, there is a strong reason to believe that terpenoids contribute to the medicinal benefits of cannabis.

There have been trials that suggest terpenoids as chemotaxonomic markers in cannabis [35]. By isolating the terpenoids of common cannabis strains sativa and indica, further insight can be gained into the therapeutic and medicinal effects of cannabis. Providing patients and health care providers with more information regarding the composition of cannabis helps them to rely on the chemical composition for reproducible physiological results. A recent study showed a large variation of the relative content of terpenes between cannabis strains and suggests that terpenoid variation can be used to characterize cannabis biotypes. Three principal terpenoids that were extracted from Cannabis sativa include beta-myrcene, limonene and alpha-pinene [34]. Each of the three strains were present in a variety of concentration on individual sativa strains. Myrcene is a recognized sedative employed to aid in sleep [34]. In a trial conducted in mice, myrcene served as a muscle relaxant and induced barbiturate sleep time at high doses [35]. Myrcene has also been found to be a strong anti-inflammatory agent and to have anti-catabolic effects in human chondrocytes, halting or slowing down cartilage destruction and osteoarthritis [36]. Limonene is also effective as a sedative agent at high doses). Limonene has been found to have anti-fungal properties [37]. The terpenoid alpha-pinene has been found to be an anti-microbial component. Positive enantiomers of alpha and beta pinene exhibit anti-microbial activity against fungi and bacteria [38].

CONCLUSION

Removal of the PHS Review by the Obama administration will initiate the next generation of research on cannabis. By permitting more federal funding and access to samples, the medicinal benefit and their bio-molecular pathways will be better understood. As state governments progressively determine the laws pertaining to cannabis legalization, federal policy makers are learning the business model to regulate and track legal cannabis, and collect tax revenues. With the availability of cannabis tracking software, full transparency of the cultivation, processing and sales of legal cannabis can be attained in real-time. Therefore, removing
cannabis from the list of Schedule II drugs and making it federally legal could lead to a significant increase in the federal funding required for research.

The medicinal benefits of cannabis have become virtually undeniable. With limited understanding of the bio-molecular composition and bio-chemical pathways that are responsible for the medicinal benefits of cannabis, further research on the ECS and cannabis is needed. The components of cannabis (THC, CBD and Terpenoids) have been analyzed and linked to the treatment of multiple conditions such as inflammation, neuro-inflammation, bone fracture healing, insomnia, cancer and epilepsy. As federal funding for cannabis research increases all over the world, a better understanding of these healing benefits will be reached.

REFERENCES