Journal of Ayurvedic and Herbal Medicine

In-vitro and *in-vivo* supportive research on medicinal properties of *Cannabis sativa*: A comprehensive review

Pathirage Kamal Perera¹, Jalathge Isurika Dilanthi Diddeniya¹

¹ Institute of Indigenous Medicine, University of Colombo, Colombo, Sri Lanka

ABSTRACT

Cannabis sativa is an herbaceous plant which is mainly used as a remedy for neurological, digestive and immunological ailments in traditional medicine. Even though Cannabis is the most illicit plant around the world, its medicinal properties are beneficial in number of ways. Numerous beneficial effects of *C. sativa* have been demonstrated in multiple *in-vitro* and *in-vivo* studies from different parts of the world. The aim of this paper was to systematically review the literature and provide a summary on potential medicinal benefits of *C. sativa*. This systematic review was conducted by using the data bases; Science direct and PubMed for studies published from 1st of January 2015 to 31st of October 2020. In order to obtain further data, a manual search was also carried out from the reference lists of included articles. After removing the duplicate articles 77 total number of articles included in this present review. The beneficial health effects of *C. sativa* were anti-inflammatory, analgesic, anti-microbial, anti-parasitic, anti-oxidant and anti-cancer properties. In addition, it revealed that *C. sativa* lower blood glucose, serum cholesterol and blood pressure levels. Apart from that, the use of Cannabis in other diseases such as irritable bowel disease, renal diseases, neurofibromatosis, and leucorrhea was also identified. The wide range of medicinal effects may be due to main active ingredients of Tetrahydro cannabinol, Cannabidiol, Cannabinol and Tetrahydro cannabivarin. Available *in-vitro* and *in-vivo* evidence suggested that *C. sativa* has many favorable health effects and further randomized controlled clinical trials will be needed to determine these effects thoroughly.

Keywords: *Cannabis sativa*, Medicinal properties, *In-vitro* and *in-vivo* studies, Health benefits.

INTRODUCTION

Cannabis sativa L. is an annual, herbaceous plant which belongs to the Cannabaceae family and it is traditionally used as a medicinal plant by different cultures around the world. Almost all parts of Cannabis plant such as tender leaves, seeds, fruits and un-pollinated inflorescence have been used for the medical and recreational purposes. The distribution of Cannabis is, in the temperate parts of Asia, Southern Siberia, Persia and probably of Northern India and China. It is cultivated in Sri Lanka, illicitly in chenas in the dry zone [1]. The active ingredients present in this plant are Tetrahydro cannabinol (THC), Cannabidiol (CBD), Cannabinol (CBN), Tetrahydro cannabivarin [2]. Due to legal restrictions implemented regarding the use, possession, cultivation, selling and distribution of Cannabis, it is considered as the most commonly used illegal drug in the world [3]. Cannabis can be used by means of smoking, vaporizing, edibles, capsules, tea and as an extract [4]. In Ayurveda medicine Cannabis is considered as a remedy for respiratory, digestive and gynecological ailments. The medical actions it possesses are digestive, appetizing, aphrodisiac, diuretic, analgesic, cough suppressing, anti-epileptic and anthelmintic actions [5]. It aids in the treatment of cough, asthma, diarrhea, dysentery, piles, neuralgia, migraine, blood poisoning [6]. The amount of THC contain in a Cannabis plant is used as a measure to identify the potency in it. Among the three main types of cannabis products named herb (marjuana), resin (hashish) and oil (hash oil), 5% of THC content is contained in flower heads and surrounding leaves while 20% of THC content is contained in Cannabis resin. Cannabis oil contain more than 60% of THC content and it is the most potent form of Cannabis [7].

Numerous beneficial health effects of *Cannabis sativa* have been identified from different parts of the world such as anti-microbial, anti-inflammatory, analgesic, reducing risk of cancer etc. The aim of this paper was to systematically review the scientific literature, arrange a summary on potential medicinal benefits of Cannabis and to conserve the precious knowledge about Cannabis. Also, to provide a guidance to researchers who wish to conduct future research based on the positive findings gathered by analyzing various researches published all around the world.

*Corresponding author:
Dr. Pathirage Kamal Perera
Senior Lecturer Gr.I, Institute of Indigenous Medicine, University of Colombo, Colombo, Sri Lanka
Email:
kamalperera@iim.cmb.ac.lk

METHODOLOGY

A systematic review was carried out by using the published studies which report about the medicinal effects of *Cannabis sativa* in accordance with PRISMA (Preferred Reporting Items for Systematic reviews and Meta-Analyses) guidelines. A comprehensive systematic review was conducted by using the data bases; Pub Med, Science direct for studies published from 1st of January, 2015 to 31st of October, 2020. The key words used in this subject were *Cannabis sativa*, medicinal properties, and health benefits. Results were obtained only from the studies published in English, while excluding commentaries and duplicate articles. The selected, appropriate articles were initially searched by reading the title and their abstracts. In this instance also, the articles which do not fulfill the requirements of inclusion criteria were excluded. Thereafter, remaining articles were screened by reading the complete text at the final stage. Those articles which did not satisfactorily enrich with inclusion criteria were excluded again.

Apart from that, relevant texts and internet sources were also used for the purpose of gathering necessary facts about Cannabis plant. This research process was conducted independently by the reviewers and the articles which were to be included in the review was determined by an iterative consensus process at the final stage.

RESULTS

Literature search

By using the above-mentioned criteria, the literature search identified these number of articles in the databases respectively; PubMed (n=122), Science direct (n=56). Seven additional articles were searched manually by searching the reference lists in order to find the possible inclusion criteria. After removing duplicate articles, the total number of articles included in the present review is 77. The search strategy is summarized in Figure 1.

In-vitro and *in-vivo* research

Anti-microbial properties

C. sativa has shown potential anti-microbial action against a wide variety of bacteria and fungi. (*Bacillus subtilis, Aliivibrio fischeri, Magnaporthe grisea, Ascosphaera apis*). There were 6 studies evaluating the anti-microbial properties. The inflorescence extracts of ten different hemp species were screened for antibacterial effects showed that strong antibacterial activities against both Gram-positive *Bacillus subtilis* and especially Gram-negative *Aliivibrio fischeri* bacteria. This effect directed analysis was conducted by using High Performance Thin Layer Chromatography (HPLC) method [8]. Findings of the study conducted by Chouhan et al. reported that silver nanoparticles (AgNPs) synthesized from *Cannabis sativa* aqueous leaf extract (CSE) exhibit anti-bacterial and anti-yeast properties [9].

Research demonstrated that C. sativa essential oil performed anti-microbial effects against tested microorganisms where this fact can be used in future to overcome the intense use of antibiotics for infectious conditions [10]. The leaf extracts from the plant *C. sativa* have reported to possess antifungal properties and can be used in treatments against the pathogen *Magnaporthe grisea* to get rid from rice blast disease [11].

Anti-microbial activities of cannabinoidal compounds which was derived by oxygenation was described [12]. Essential oil of *C. sativa* known as hemp seed oil was identified as minimally effective in giving out the anti-fungal property against *Ascosphaera apis* who is one of the major fungal pathogens of honey bee broods and the causative agent of Chalk brood disease. A study was conducted for evaluating *in vitro* antifungal activities of 27 plant essential oils against two isolates of *A. apis* (Aksu-4 and Aksu-9) [13].
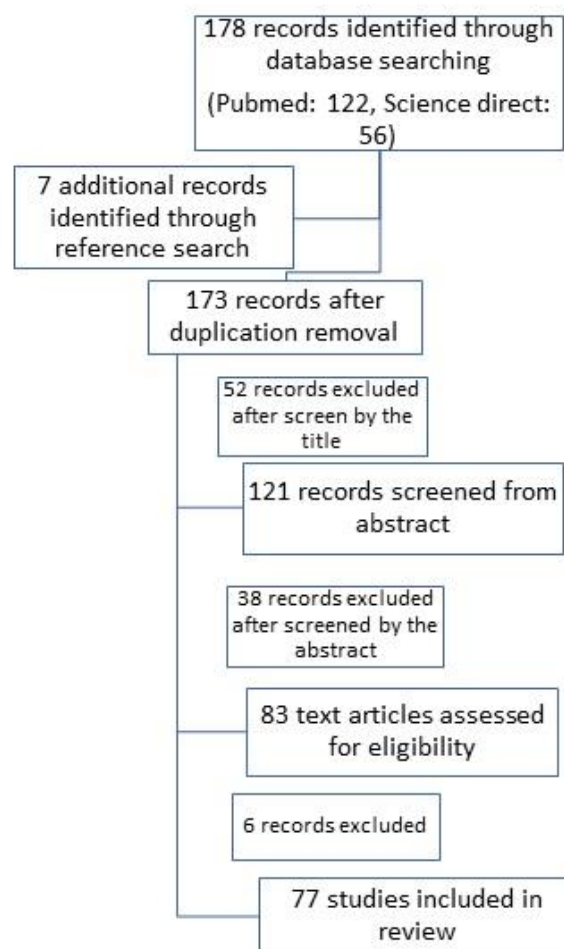

Figure 1: Summarized search strategy

Anti-parasitic effects

Rossi et al. evaluated the mosquitocidal activities of essential oils obtained from different hemp varieties on larvae and pupae of two main malaria vectors, *Anopheles gambiae* and *An. Stephensi* [14]. Antileishmanial activities of *C.sativa* was evaluated and it has been mentioned that the results obtained can be used in futute antileishmanial drug development [15]. Research demonstrated that there is anti-malarial and antileishmanial activities in cannabinoid compounds tested by oxygenation of them. Essential oils of C. sativa was evaluated [12] and their acute toxicity was assessed against the invasive species; mosquito *Aedes albopictus* and, the freshwater bladder snail *Physella acuta* by *Stefano* et al. [16].

Effects on blood pressure, glycaemia control and lipids

Beneficial effects of cannabis sativa extracts on the initial stage of atherosclerosis was identified by means of characterizing the chemical

profile of three varieties of *C. sativa* available in Uruguay [17]. *Trentin* et al. tested the hypothesis that Cannabidiol (CBD) treatment improves metabolic dysfunctions in middle-aged diabetic rats submitted to chronic cerebral hypo perfusion [18]. The findings revealed that CBD reduced hyperglycemia and it increased the insulin secretion as well as it lowered the total cholesterol level giving out a hepatoprotective effect. Δ9-THCA-A reduce body fat mass and liver steatosis in HFD fed mice as well as it improves glucose tolerance, insulin sensitivity and insulin profiles *in vivo* were experimented [19]. A study revealed that combination of the fast-acting hemp seed protein hydrolysates (HPH) (1% alcalase) with the longer-lasting HPHs (2% and 4% pepsin) could provide daily effective systolic blood pressure reductions. It was observed by administration of HPH orally to hypertensive rats and observing their pressure reduction for twenty-four hours [20].

Anti-oxidant properties

By making use of hypercholesterolemic rats, demonstrated that anti-oxidant effect of hemp could alleviate pathological conditions such as fatty liver disease [21]. Antioxidant property of two different non drug *C. sativa* essential oils by several in vitro cell-free assays and it was shown that they possessed strong antioxidant properties [22]. The total antioxidant capacity determined *in vitro* by phosphomolybdenum assay has exhibited Cannabis contain antioxidants [23]. Radical scavenging activity of 16 plants extracts harvested in South Hungary was assessed by means of DPPH (1, 1-diphenyl-2-picrylhydrazyl) assay [24] has investigated that *C.sativa* extract possess remarkable anti-oxidant activity among other plants. The experimental findings have suggested that *C. sativa* essential oil can be considered as a potential source of natural antioxidant [12].

The antioxidant effect of *C. sativa* L. seeds and sprouts was evaluated and their results showed that antioxidant activity was higher in sprouts than in seeds of *C. sativa* [25]. Zimniewska et al. tested the antioxidant properties of flax and hemp fibers with the use of FRAP and DPPH methods and identified that anti-oxidant potential of hemp and flax fibers depend on their chemical composition [26]. *In* vitro antioxidant activity of antioxidant enzymes in HepG2 cells by using Ethanol and supercritical fluid (SF) extracts obtained from de-hulled hemp seed and demonstrated that the hemp seed effectively inhibited H_2O_2 mediated oxidative stress [27]. Furthermore, it has been stated that it is effective for revealing the diseases mediated by oxidative stress.

Anticonvulsant effects

It has shown Cannabidiol, which is a constituent of *Cannabis sativa* prevented cocaine-induced seizures in mice [28]. The anti-epileptic effects of certain Cannabis compounds were tested by using zebra fish, five different cannabis compounds and they were compared in both chemically-induced and Dravet Syndrome epilepsy models [29]. *Royston* et al. carried out an experiment and the collected data highlighted that transient receptor potential vanilloid 1 (TRPV1) has the potential mechanisms of action for the anticonvulsive effects of Cannabidiol (CBD) [30]. An online survey was conducted among United States based neurologists, nurse practitioners/ nurses, and pharmacists observed that use of medical cannabis was highly familiar among them [31]. It has reported the use of medical marijuana (MM) in the treatment of epilepsy among pediatric population [32]. Also, it has mentioned the risks and benefits of MM in pediatric epilepsy. The findings demonstrated that there was a significant reduction in seizure behavior. From an ethnobotanical survey demonstrated that quantitative analyses Informant consensus factor (ICF), Relative frequency citation (RFC), Use value (UV), Fidelity level (FL), Relative importance (RI) of documented data revealed the medicinal uses of *C. sativa* such as it aids in convulsions, diarrhea, indigestion and obesity [33].

Anti-cancer properties

Buchwald et al. carried out a qualitative interview study among cancer patients and identified that, the use of cannabis-based medicine (CBM) as a supplement to conventional palliative care [34]. The extracts of *C. sativa* were used to assess the cytotoxic effect against human colon cancer cells and it was shown that these extracts possess direct selective cytotoxic effect on colon cells [35]. Further, it was stated that extracts of *C. sativa* could reverse the drug resistance in colorectal carcinoma cells. It was examined the *in vitro* effects of CBD on human gastric cancer SGC-7901 cells and suggested that CBD have therapeutic effects on gastric cancer [36]. The effect of cannabinoids on tumor growth *in vivo* was studied in a zebrafish xenograft model [37]. The *in vitro* and *in vivo* study proven that Cannabidiol have anti-neoplastic properties and enhance the immune response to cancer [38]. *Osman* et al. demonstrated that eighteen new compounds of oxygenated cannabinoids has shown cytotoxic activity against cancer cells by testing these compounds for their modulatory effect on cannabinoid receptors CB1 and CB2 [39]. Ammar et al. evaluated in-vitro anti-proliferative activities of endocannabinoid reuptake inhibitor OMDM-2 alone /in combination with curcumin in breast cancer and synergism was identified [40]. Further *Thomas* et al. evaluated the incidence of bladder cancer among the men enrolled in California Men's Health study by giving questionnaires and their findings demonstrated that, the use of Cannabis among men has significantly a decreased risk of bladder cancer [41].

Anti-inflammatory effects

Noelia et al. investigated chronic inflammatory conditions were managed by hemp protein products (HPPs) and promote regenerative processes by means of reprogramming monocytes toward M2 polarization phenotype [42]. The effects and action of cannabinoid receptor agonists, including Δ9-THC on inflammation and organ injury was investigated in endotoxemic mice [43]. A study carried out by Kaushal et al. suggested that anti-inflammatory property of hemp was useful in alleviating pathophysiological conditions including fatty liver disease [21]. Rossi et al. evaluated the anti-inflammatory effect of different hemp varieties and it was observed that appropriate doses of hemp exerted an anti-inflammatory effect on human cell lines 14]. *Majdi* et al. discussed the possible role of cannabidiol (CBD) and delta–9–tetrahydrocannabinol (Δ⁹–THC), as a potent an anti-inflammatory agent in the treatment of (methamphetamine) METH-induced neuro-inflammation [44]. Cannabidiol (CBD) has a differential inflammatory response and acts as an anti-inflammatory agent in pro-inflammatory conditions by treating epithelial cells (BEAS-2B and NHBE), macrophages (U937), and lung fibroblast cells (HFL-1) with varying CBD concentrations or exposed to CBD aerosols [45].

Essential oils made from three monoecious nonpsychoactive chemotypes of Cannabis were analyzed for their terpenoid content and

pharmacological studies were carried out for their anti-inflammatory properties *in vitro* and *in vivo* demonstrated that terpenoids in Cannabis can be used in acute inflammatory conditions [46]. Thirteen anti-inflammatory compounds were characterised and filtered out from medicinal plant species; *Cannabis sativa, Prunella vulgaris* and *Withania somnifera* and analysed them for Rheumatoid Arthritis by targeting TNF-α through in silico analyses [47]. This study revealed that these plants possess anti-inflammatory, anti-arthritic and anti-rheumatic properties. The immune-cell modulatory properties of medical cannabis were investigated and stated that it can be used in the treatment of anti-inflammatory conditions [48].

In-vitro and in-vivo analgesic properties

A descriptive study was carried out by means of questionnaire sent to dronabinol (a pure extract of delta-9-tetrahydrocannabinol) prescribers throughout metropolitan France identified that it aids in the management of pain [49]. *Kevin* et al. examined the Cannabis use preferences among medical cannabis users with chronic pain from an ongoing online survey by using 1321 participants [50]. By conducting an electronic survey in medicinal cannabis patients with headache, arthritis, and chronic pain, it was revealed that chronic pain was the most common reason for cannabis used [51]. The findings of *Nicole* et al. showed that HUF-101, a fluorinated CBD analogue produced antinociceptive effects at lower doses than CBD, indicating that the addition of fluoride improved its pharmacological profile which induces pain relief [52].

Neuro-protective effects

An optical coherence tomography (OCT) study demonstrated that increase in retinal nerve fiber layer thickness represent the neuroprotective effect of Cannabis [53]. *Stephanie* et al. [54] conducted a randomized, double-blinded, placebo-controlled, crossover clinical trial by using twenty four individuals and evaluated the impacts of acute Cannabidiol (CBD) administration (a dose of 300 mg) on anxiety and tremors induced by a Simulated Public Speaking Test (SPST) in people with Parkinson's disease (PD). It was observed that acute CBD administration decreased anxiety in patients with PD, and there was also decreased tremor amplitude. The ability of Cannabis to lower the neurocognitive impairment in HIV infected people was examined by using 679 people living with HIV and 273 people living without HIV [55]. Antonella et al. evaluated the neuroactive effects of two different types of *C. sativa* essential oils on mouse cortical neuronal and also on human iPS cell-derived central nervous system cells grown on MEA chips [56].

A retrospective analysis was performed on a cohort of 146 medical cannabis patients who reported benzodiazepine use at initiation of cannabis therapy indicated that 45.2% patients successfully discontinued their pre-existing benzodiazepine therapy [57]. It demonstrated the use of medical Cannabis in giving sedative properties for anxiety and other neurological conditions. *In-vitro* neuroprotective properties of *C. sativa* was evaluated by testing the novel cannabis flavonoid, cannflavin; indicated that there was a concentration dependent neuroprotective effect of Cannabis [58]. An experiment was carried out in male rodents and observed that CBD could induce rapid and sustained antidepressant-like effects relevant for depression [59]. *Lejczak* et al. demonstrated the use of dronabinol (a pure extract of

delta-9-tetrahydrocannabinol) in the management of multiple sclerosis and Parkinson's disease [49]. Neuroprotective role of cannabinoid derivate such as cannabidiol (CBD) and delta–9–tetrahydrocannabinol (Δ9–THC) in multiple neurodegenerative diseases has been discussed [44]. The efficacy of Cannabivarin (CBDV) for Rett syndrome (RTT) which is a rare neurodevelopmental disorder was evaluated by using rats demonstrated that, the used treatment could restore the compromised health condition [60,61]. *Brishna* et al. carried out a survey in order to identify the effectiveness of Cannabis for treating anxiety and it was recorded that cannabis is highly effective for treating anxiety with an average score of 8.03 on a Likert scale of 0 to 10 (0 = not effective, 10 = extremely effective).

Guan et al. performed gene microarray analysis in order to verify the key regulatory function of Homeodomain-interacting protein kinase 2 (HIPK2) in Parkinson's disease (PD). (*Z*)-methyl *p*-hydroxycinnamate (ZMHC, 7) with other eighteen compounds were isolated from *Cannabis sativa* subsp. *sativa*, and stated that ZMHC inhibited HIPK2, which attenuated MPP$^+$-induced apoptosis in SH-SY5Y cells. It was observed that ZMHC could be a potential precursor agent for future PD therapy [62]. The effect of chronic CBD treatment on cognition and social interaction demonstrated by using rats and it was revealed that CBD can be used in treating the cognitive deficits and social withdrawal of schizophrenia [63].

Justin et al. demonstrated that therapeutic properties of Cannabis can be made use for the treatment of schizophrenia-related psychoses [64]. The experiment was conducted by using rats *in vivo* has stated that Cannabinoids could avoid the effects of inflammation in order to create a reparative environment in neurodegenerative diseases such as Alzheimer's disease, multiple sclerosis [65].

Ester et al. demonstrated that there was preserved memory in AβPP/PS1 transgenic mice when delta-9-tetrahydrocannabinol (THC) or cannabidiol (CBD) botanical extracts were chronically administered during early symptomatic stage and combination of THC and CBD exhibits a better therapeutic profile than using each cannabis component alone which support the use of a cannabis-based medicine as potential therapy against Alzheimer's disease [66]. Results obtained by *Campos* et al. [by treating the female mice with Cannabidiol (CBD) indicated that CBD exhibits neuroprotective effects in Cerebral Malaria model [67].

Neuroprotective properties of CBD were demonstrated by evaluating the animal models and it was revealed that inhibition of microglial activation improve schizophrenia symptoms [68]. Malomo et al. investigated the *in vitro* pre-treatment of mesenchymal stromal cells (MSCS) obtained from human gingiva (hGMSCs) with Cannabidiol (CBD) and stated that human gingiva-derived MSCs conditioned with CBD could represent a valid method for improving the hGMSCs phenotype and it might be a potential therapeutic tool in the treatment of neurodegenerative diseases [20].

Other medicinal properties

Firouzi et al. conducted an experiment in mice to evaluate the effects of evening primrose/hemp seed oil (EPO/HSO) on improving the membrane fatty acids composition of spleen and blood cells and immunologic factors in compared to rapamycin (RAPA) in autoimmune

encephalomyelitis [69]. Therapeutic potential of Δ^8–Tetrahydrocannabivarin (Δ^8–THCV) for the treatment of nicotine dependence in rodents were observed [70]. A total of 128 individuals with chronic pain who were above 50 years of age were selected from the *Rambam* Institute for Pain Medicine in Haifa, Israel examined the association between use of cannabis and sleep problems among chronic pain patients. It was revealed that there was positive effect of Cannabis use on sleep maintaining in chronic pain patients [71]. A qualitative evaluation was conducted by utilizing a focus group, semi structured interviews and a community event observed that Cannabis-Derived Medicinal Products (CDMP) in the Treatment of acute postoperative pain, nausea, and vomiting [72]. Bhatt et al. conducted a randomly assigned experiment by using male and female Sprague-Dawley rats suggested that $\Delta 9$-tetrahydrocannabinol (THC) has potential therapeutic benefit on post-concussive symptomology in mild traumatic brain injury [73].

Omar et al. reported the use of Cannabidiol (CBD) for the management of chronic pain and concomitant mood disorder in the common genetic disorder named; Neurofibromatosis (NF1) by conducting a case report [74]. Another study proposed that CB_2 agonists as potential drugs for development of novel therapeutic modality to traumatic brain injury and it was demonstrated by observing the enhanced neurobehavioral recovery in mice and rats subjected to closed-head injury [75]. An ethno-gynecological survey was performed in a tribal region near the Pak-Afghan border has reported that fifty-two medicinal plants including *C. sativa* has been used for the treatment of numerous gynecological disorders such as leucorrhea, amenorrhea, uterine infection etc. Especially, *C. sativa* has been used in the treatment of abdominal pain, gonorrhea, pregnancy and female impotency in the forms of powder with water [76]. *Singh* et al. examined the use of Cannabis in dermatology even though it has been categorized in the dangerous plant category [77].

Usage of medical marijuana in the end stage renal disease (ESRD) and chronic kidney disease (CKD) has been described [78]. The use of Cannabis in the treatment of pediatric Inflammatory Bowel Disease (IBD) was identified [79]. Androvicova et al. conducted an open-randomized study with 21 heterosexual casual cannabis users and observed the effect of Cannabis in low sexual desires [80].

Herbal medicines used in the treatment of Tuberculosis by Traditional medical practitioners of the eastern region of O.R. Tambo district was identified. Among them Twenty-four plant species belonging to 19 families that are used for the treatment of Tuberculosis including *C. sativa* was reported. Moreover, it stated that leaf and root are the used parts of the plant and it can be given by orally or infusion [81]. An ethno botanical survey was carried out among the Gbaya ethnic group of Bertoua city in the East region of Cameroon demonstrated that there were several plant species which are used as traditional cosmetics and among them Cannabis is used in the hair care treatment [82]. In the article published by *Adam* et al. named "Complementary and Alternative Medicines Used by Patients with Inflammatory Bowel Diseases (IBD)" has been stated that there were evidence for the effects of herbs such as *C. sativa* in the treatment of IBD [83].

Natalya et al. conducted an experiment by giving a mixture of CBD and Δ (9) -tetrahydrocannabinol (THC) for eight weeks for rats and the data showed that major non-psychoactive cannabis constituent, cannabidiol (CBD) leads to improvement in fracture healing [84]. Yeshurun et al. conducted a phase II study and concluded that of combination of Cannabidiol (CBD) with Graft-versus-host-disease (GVHD) prophylaxis was a safe method to reduce the incidence of acute GVHD [85]. Sleep was induced in rats by means of vaporized cannabis (administration of low doses of THC) has shown increment of NREM sleep, but only during the light (resting) phase [86].

DISCUSSION

The available in-vitro and in-vivo studies suggest that, *C. sativa* has anti-microbial, anti-parasitic, anti-oxidant, anti-inflammatory, anti-cancer and analgesic properties. In addition, it seems to lower blood glucose, serum cholesterol and blood pressure levels. The different parts of *C. sativa* plant possess different amounts of active ingredients in it and this diversity of chemical constituents is likely to be the reason for the wide range of medicinal effects observed. Further identifications of such mechanisms responsible for these medicinal effects would be interesting to follow.

It was noted that, leaf extracts of Cannabis as well as essential oils are responsible for giving the anti-microbial activities against microbes and for giving anti-parasitic effects too. An active ingredient in C. sativa, named "Cannabidiol" (CBD) was likely to responsible for giving anti-convulsive properties. Further studies are needed to be carried out with regard to this for further clarification. The mechanism of action by which *C. sativa* reduces blood glucose level has been well studied *in-vitro* and *in-vivo*. It seems that *C. sativa* a) increases the insulin secretion, b) improves glucose tolerance and c) improves insulin sensitivity. Hemp seed protein hydrolysates (HSPH) provided the effect of lowering the systolic blood pressure but the exact blood pressure reducing mechanism and lipid lowering mechanism was not clearly mentioned. Cannabidiol (CBD) was the key factor for giving anti-cancer effects by giving actions such as a) cytotoxic activity against cancer cells, b) anti-neoplastic properties and c) enhance the immune response against cancers. Delta-9-tetrahydrocannabinol (THC) or Cannabidiol (CBD) were mainly beneficial in giving the neuroprotective effects and anti-inflammatory effects.

CONCLUSION

The available in-vitro and in-vivo studies suggested that *C. sativa* has anti-microbial, anti-parasitic, anti-oxidant, anti-inflammatory, anti-cancer, analgesic and neuroprotective properties. Apart from that it possesses the actions of reducing blood glucose, serum cholesterol and blood pressure levels too. Further *C. sativa* was aid in alleviating other disease conditions as well. Some of them were IBD, renal diseases, neurofibromatosis etc. However randomized controlled clinical trial will be necessary to determine whether above effects have community health implications.

Data availability

The data used to support the findings of this study are available from the corresponding author upon request.

Conflict of Interest

None declared.

REFERENCE

1. Jayaweera DMA, Lilani KS. Medicinal plants (Indigenous and Exotic) used in Ceylon. Part 2. Colombo: The National Science foundation Sri Lanka; 2006. 6-7 p.

2. Atakan Z. Cannabis, a complex plant: different compounds and different effects on individuals. Therapeutic advances in psychopharmacology. 2012;2(6):241-54.

3. Cannabis: uses and effectiveness. Web med. Available from: https://www.webmd.com/vitamins/ai/ingredientmono-947/marijuana.

4. Cannabis in Sri Lanka: Laws, use and history. (2021). Available from: https://sensiseeds.com/en/blog/countries/cannabis-in-sri-lanka-laws-use-history/.

5. Cannabis use in Sri Lanka. National council for mental health. Available from : http://www.ncmh.lk/page/cannabis-use-in-sri-lanka/99

6. Department of Ayurveda, Ayurveda pharmacopoeia: Vol. 1- part 111, 2nd ed, 77-78 p.

7. Jayaweera DMA, Lilani KS. Medicinal plants (Indigenous and Exotic) used in Ceylon. Part 2. Colombo: The National Science foundation Sri Lanka; 2006. 6-7 p.

8. Why does Cannabis potency Matter? United Nations office on Drugs and Crime. Available from www.unodc.org/unodc/en/frontpage/2009/June

9. Corni G, Brighenti V, Pellati F, Morlock GE. Effect-directed analysis of bioactive compounds in *Cannabis sativa* L. by high-performance thin-layer chromatography. Journal of Chromatography A. 2020;1629:461511.

10. Chouhan S, Guleria S. Green synthesis of AgNPs using Cannabis sativa leaf extract: Characterization, antibacterial, anti-yeast and α-amylase inhibitory activity. Materials Science for Energy Technologies. 2020;3:536-44.

11. Nafis A, Kasrati A, Jamali CA, Mezrioui N, Setzer W, Abbad A et al. Antioxidant activity and evidence for synergism of *Cannabis sativa* (L.) essential oil with antimicrobial standards. Industrial Crops and Products. 2019;137:396-400.

12. Dar MS, Ganaie SA, Raja W, Teeli RA. *In-vivo* investigation on antifungal properties of leaf extracts of certain medicinal plants through seed treatment and foliar sprays against rice blast disease (*Magnaporthe grisea*) in Kashmir, India. Annals of Agrarian Science. 2018;16(3):267-71.

13. Osman AG, Elokely KM, Yadav VK, Carvalho P, Radwan M, Slade D et al. Bioactive products from singlet oxygen photooxygenation of cannabinoids. European Journal of Medicinal Chemistry. 2018;143:983-96.

14. Ansari MJ, Al-Ghamdi A, Usmani S, Khan KA, Alqarni AS, Kaur M et al. *In vitro* evaluation of the effects of some plant essential oils on *Ascosphaera apis*, the causative agent of Chalkbrood disease. Saudi journal of biological sciences. 2017;24(5):1001-6.

15. Rossi P, Cappelli A, Marinelli O, Valzano M, Pavoni L, Bonacucina G et al. Mosquitocidal and anti-inflammatory properties of the essential oils obtained from monoecious, male, and female inflorescences of hemp (*Cannabis sativa* L.) and their encapsulation in nanoemulsions. Molecules. 2020;25(15):3451.

16. Mehwish S, Islam A, Ullah I, Wakeel A, Qasim M, Khan MA et al. *In vitro* antileishmanial and antioxidant potential, cytotoxicity evaluation and phytochemical analysis of extracts from selected medicinally important plants. Biocatalysis and agricultural biotechnology. 2019;19:101117.

17. Bedini S, Flamini G, Cosci F, Ascrizzi R, Benelli G, Conti B. *Cannabis sativa* and *Humulus lupulus* essential oils as novel control tools against the invasive mosquito *Aedes albopictus* and fresh water snail *Physella acuta*. Industrial Crops and Products. 2016;85:318-23.

18. Musetti B, González-Ramos H, González M, Bahnson EM, Varela J, Thomson L. *Cannabis sativa* extracts protect LDL from Cu2+-mediated oxidation. Journal of cannabis research. 2020;2(1):1-1.

19. Zorzenon MR, Santiago AN, Mori MA, Piovan S, Jansen CA, Padilha ME et al. Cannabidiol improves metabolic dysfunction in middle-aged diabetic rats submitted to a chronic cerebral hypoperfusion. Chemico-biological interactions. 2019;312:108819.

20. Palomares B, Ruiz-Pino F, Garrido-Rodriguez M, Prados ME, Sánchez-Garrido MA, Velasco I et al. Tetrahydrocannabinolic acid A (THCA-A) reduces adiposity and prevents metabolic disease caused by diet-induced obesity. Biochemical Pharmacology. 2020;171:113693.

21. Malomo SA, Onuh JO, Girgih AT, Aluko RE. Structural and antihypertensive properties of enzymatic hemp seed protein hydrolysates. Nutrients. 2015;7(9):7616-32.

22. Kaushal N, Gupta M, Kulshreshtha E. Hempseed (*Cannabis sativa*) lipid fractions alleviate high-fat diet-induced fatty liver disease through regulation of inflammation and oxidative stress. Heliyon. 2020;6(7):e04422.

23. Smeriglio A, Trombetta D, Alloisio S, Cornara L, Denaro M, Garbati P et al. Promising *in vitro* antioxidant, anti-acetylcholinesterase and neuroactive effects of essential oil from two non-psychotropic *Cannabis sativa* L. biotypes. Phytotherapy Research. 2020;34(9):2287-302.

24. Mehwish S, Islam A, Ullah I, Wakeel A, Qasim M, Khan MA et al. *In vitro* antileishmanial and antioxidant potential, cytotoxicity evaluation and phytochemical analysis of extracts from selected medicinally important plants. Biocatalysis and agricultural biotechnology. 2019;19:101117.

25. Szerlauth A, Muráth S, Viski S, Szilagyi I. Radical scavenging activity of plant extracts from improved processing. Heliyon. 2019;5(11):e02763.

26. Frassinetti S, Moccia E, Caltavuturo L, Gabriele M, Longo V, Bellani L et al. Nutraceutical potential of hemp (*Cannabis sativa* L.) seeds and sprouts. Food chemistry. 2018;262:56-66.

27. Zimniewska M, Rozańska W, Gryszczynska A, Romanowska B, Kicinska-Jakubowska A. Antioxidant potential of hemp and flax fibers depending on their chemical composition. Molecules. 2018;23(8):1993.

28. Hong S, Sowndhararajan K, Joo T, Lim C, Cho H, Kim S et al. Ethanol and supercritical fluid extracts of hemp seed (*Cannabis sativa* L.) increase gene expression of antioxidant enzymes in HepG2 cells. Asian Pacific Journal of Reproduction. 2015;4(2):147-52.

29. Gobira PH, Vilela LR, Gonçalves BD, Santos RP, de Oliveira AC, Vieira LB et al. Cannabidiol, a Cannabis sativa constituent, inhibits cocaine-induced seizures in mice: possible role of the mTOR pathway and reduction in glutamate release. Neurotoxicology. 2015 Sep 1;50:116-21.

30. Thornton C, Dickson KE, Carty DR, Ashpole NM, Willett KL. Cannabis constituents reduce seizure behavior in chemically-induced and scn1a-mutant zebrafish. Epilepsy & Behavior. 2020;110:107152.

31. Gray RA, Stott CG, Jones NA, Di Marzo V, Whalley BJ. Anticonvulsive properties of cannabidiol in a model of generalized seizure are transient receptor potential vanilloid 1 dependent. Cannabis and Cannabinoid Research. 2020;5(2):145-9.

32. Jeyaraj MK, Menon RN, Justus S, Alexander A, Sarma PS, Radhakrishnan K. A critical evaluation of the lateralizing significance of material-specific memory deficits in patients with mesial temporal lobe epilepsy with hippocampal sclerosis. Epilepsy & Behavior. 2013;28(3):460-6.

33. Markle M, Nativio DG. Medical Marijuana in the Pediatric Population With Epilepsy—What You Should Know. Journal of Pediatric Health Care. 2019;33(6):626-32.

34. Rehman MN, Ahmad M, Sultana S, Zafar M, Edwards S. Relative popularity level of medicinal plants in Talagang, Punjab Province, Pakistan. Revista Brasileira de Farmacognosia. 2017;27:751-75.

35. Buchwald D, Brønnum D, Melgaard D, Leutscher PD. Living with a hope of survival is challenged by a lack of clinical evidence: an interview study among cancer patients using cannabis-based medicine. Journal of Palliative Medicine. 2020;23(8):1090-3.

36. Mangoato IM, Mahadevappa CP, Matsabisa MG. *Cannabis sativa* L. Extracts can reverse drug resistance in colorectal carcinoma cells *in vitro*. Synergy. 2019;9:100056.

37. Zhang X, Qin Y, Pan Z, Li M, Liu X, Chen X et al. Cannabidiol induces cell cycle arrest and cell apoptosis in human gastric cancer SGC-7901 cells. Biomolecules. 2019;9(8):302.

38. Tomko A, O'Leary L, Trask H, Achenbach JC, Hall SR, Goralski KB et al. Antitumor activity of abnormal cannabidiol and its analog O-1602 in taxol-resistant preclinical models of breast cancer. Frontiers in Pharmacology. 2019:1124.

39. Sulé-Suso J, Watson NA, van Pittius DG, Jegannathen A. Striking lung cancer response to self-administration of cannabidiol: A case report and literature review. SAGE Open Medical Case Reports. 2019;7:2050313X19832160.

40. Osman AG, Elokely KM, Yadav VK, Carvalho P, Radwan M, Slade D et al. Bioactive products from singlet oxygen photooxygenation of cannabinoids. European Journal of Medicinal Chemistry. 2018 Jan 1;143:983-96.

41. Ammar RM, Ulrich-Merzenich G. *Curcumin synergizes* with the endocannabinoid reuptake inhibitor OMDM-2 in human MCF-7 breast cancer and U-87 glioblastoma cells. Synergy. 2017;5:7-14.

42. Li Z, Zhu Y, Kong C, Yin L, Gao Z, Zhao W, Gong D. Corticotropin-independent Cushing's syndrome in patients with bilateral adrenal masses. Urology. 2011;77(2):417-21.

43. Rodriguez-Martin NM, Montserrat-de la Paz S, Toscano R, Grao-Cruces E, Villanueva A, Pedroche J et al. Hemp (*Cannabis sativa* L.) protein hydrolysates promote anti-inflammatory response in primary human monocytes. Biomolecules. 2020 May;10(5):803.

44. Joffre J, Yeh CC, Wong E, Thete M, Xu F, Zlatanova I et al. Activation of CB1R promotes lipopolysaccharide-induced IL-10 secretion by monocytic myeloid-derived suppressive cells and reduces acute inflammation and organ injury. The Journal of Immunology. 2020;204(12):3339-50.

45. Majdi F, Taheri F, Salehi P, Motaghinejad M, Safari S. Cannabinoids Δ9-tetrahydrocannabinol and cannabidiol may be effective against methamphetamine induced mitochondrial dysfunction and inflammation by modulation of Toll-like type-4 (Toll-like 4) receptors and NF-κB signaling. Medical Hypotheses. 2019;133:109371.

46. Muthumalage T, Rahman I. Cannabidiol differentially regulates basal and LPS-induced inflammatory responses in macrophages, lung epithelial cells, and fibroblasts. Toxicology and applied pharmacology.2019;382:114713.

47. Gallily R, Yekhtin Z, Hanuš LO. The anti-inflammatory properties of terpenoids from cannabis. Cannabis and cannabinoid research. 2018;3(1):282-90.

48. Zaka M, Sehgal SA, Shafique S, Abbasi BH. Comparative in silico analyses of *Cannabis sativa, Prunella vulgaris* and *Withania somnifera* compounds elucidating the medicinal properties against rheumatoid arthritis. Journal of Molecular Graphics and Modelling. 2017;74:296-304.

49. Utomo WK, De Vries M, Braat H, Bruno MJ, Parikh K, Comalada M, Peppelenbosch MP, Van Goor H, Fuhler GM. Modulation of human peripheral blood mononuclear cell signaling by medicinal cannabinoids. Frontiers in molecular neuroscience. 2017 Jan 24;10:14.

50. Lejczak S, Rousselot H, Di Patrizio P, Debouverie M. Dronabinol use in France between 2004 and 2017. Revue Neurologique. 2019 May 1;175(5):298-304.

51. Boehnke KF, Scott JR, Litinas E, Sisley S, Clauw DJ, Goesling J, Williams DA. Cannabis use preferences and decision-making among a cross-sectional cohort of medical cannabis patients with chronic pain. The Journal of Pain. 2019 Nov 1;20(11):1362-72.

52. Baron EP, Lucas P, Eades J, Hogue O. Patterns of medicinal cannabis use, strain analysis, and substitution effect among patients with migraine, headache, arthritis, and chronic pain in a medicinal cannabis cohort. The journal of headache and pain. 2018 Dec;19(1):1-28.

53. Silva NR, Gomes FV, Fonseca MD, Mechoulam R, Breuer A, Cunha TM, Guimaraes FS. Antinociceptive effects of HUF-101, a fluorinated cannabidiol derivative. Progress in Neuro-Psychopharmacology and Biological Psychiatry. 2017 Oct 3;79:369-77.

54. Kalenderoglu A, Orum MH, Karadag AS, Kustepe A, Celik M, Egilmez OB, Eken-Gedik D. Increases in retinal nerve fiber layer thickness may

55. represent the neuroprotective effect of cannabis: an optical coherence tomography study. Journal of addictive diseases. 2020 Apr 2;38(3):280-90.

55. de Faria SM, de Morais Fabrício D, Tumas V, Castro PC, Ponti MA, Hallak JE, Zuardi AW, Crippa JA, Chagas MH. Effects of acute cannabidiol administration on anxiety and tremors induced by a Simulated Public Speaking Test in patients with Parkinson's disease. Journal of Psychopharmacology. 2020 Feb;34(2):189-96.

56. Watson CW, Paolillo EW, Morgan EE, Umlauf A, Sundermann EE, Ellis RJ, Letendre S, Marcotte TD, Heaton RK, Grant I. Cannabis exposure is associated with a lower likelihood of neurocognitive impairment in people living with HIV. Journal of acquired immune deficiency syndromes (1999). 2020 Jan 1;83(1):56.

57. Smeriglio A, Trombetta D, Alloisio S, Cornara L, Denaro M, Garbati P, Grassi G, Circosta C. Promising in vitro antioxidant, anti-acetylcholinesterase and neuroactive effects of essential oil from two non-psychotropic Cannabis sativa L. biotypes. Phytotherapy Research. 2020 Sep;34(9):2287-302.

58. Purcell C, Davis A, Moolman N, Taylor SM. Reduction of benzodiazepine use in patients prescribed medical cannabis. Cannabis and Cannabinoid Research. 2019 Sep 1;4(3):214-8.

59. Eggers C, Fujitani M, Kato R, Smid S. Novel cannabis flavonoid, cannflavin A displays both a hormetic and neuroprotective profile against amyloid β-mediated neurotoxicity in PC12 cells: Comparison with geranylated flavonoids, mimulone and diplacone. Biochemical Pharmacology. 2019 Nov 1;169:113609.

60. Sales AJ, Fogaça MV, Sartim AG, Pereira VS, Wegener G, Guimarães FS, Joca SR. Cannabidiol induces rapid and sustained antidepressant-like effects through increased BDNF signaling and synaptogenesis in the prefrontal cortex. Molecular neurobiology. 2019 Feb;56(2):1070-81.

61. Vigli D, Cosentino L, Raggi C, Laviola G, Woolley-Roberts M, De Filippis B. Chronic treatment with the phytocannabinoid Cannabidivarin (CBDV) rescues behavioural alterations and brain atrophy in a mouse model of Rett syndrome. Neuropharmacology. 2018 Sep 15;140:121-9.

62. Kamal BS, Kamal F, Lantela DE. Cannabis and the anxiety of fragmentation—a systems approach for finding an anxiolytic cannabis chemotype. Frontiers in Neuroscience. 2018:730.

63. Wang G, Zhu L, Zhao Y, Gao S, Sun D, Yuan J et al. A natural product from *Cannabis sativa* subsp. sativa inhibits homeodomain-interacting protein kinase 2 (HIPK2), attenuating MPP+-induced apoptosis in human neuroblastoma SH-SY5Y cells. Bioorganic Chemistry. 2017;72:64-73.

64. Lecca S, Melis M, Luchicchi A, Muntoni AL, Pistis M. Inhibitory inputs from rostromedial tegmental neurons regulate spontaneous activity of midbrain dopamine cells and their responses to drugs of abuse. Neuropsychopharmacology. 2012;37(5):1164-76.

65. Renard J, Loureiro M, Rosen LG, Zunder J, De Oliveira C, Schmid S et al. Cannabidiol counteracts amphetamine-induced neuronal and behavioral sensitization of the mesolimbic dopamine pathway through a novel mTOR/p70S6 kinase signaling pathway. Journal of Neuroscience. 2016;36(18):5160-9.

66. Mecha M, Carrillo-Salinas FJ, Feliú A, Mestre L, Guaza C. Microglia activation states and cannabinoid system: Therapeutic implications. Pharmacology & therapeutics. 2016;166:40-55.

67. Aso Pérez E, Sànchez ÀS, Vegas Lozano E, Maldonado R, Ferrer IF. Cannabis-based medicine reduces multiple pathological processes in AβPP/PS1 mice. Journal of Alzheimer's Disease, 2015, vol. 43, num. 3, p. 977-991.

68. Campos AD, Brant F, Miranda AS, Machado FS, Teixeira AL. Cannabidiol increases survival and promotes rescue of cognitive function in a murine model of cerebral malaria. Neuroscience. 2015;289:166-80.

69. Jarskog LF, Lowy MT, Grove RA, Keefe RS, Horrigan JP, Ball MP et al. A Phase II study of a histamine H3 receptor antagonist GSK239512 for cognitive impairment in stable schizophrenia subjects on antipsychotic therapy. Schizophrenia research. 2015;164(1-3):136-42.

70. Rezapour-Firouzi S, Mohammadian M, Sadeghzadeh M, Mazloomi E. Effects of co-administration of rapamycin and evening primrose/hemp seed oil supplement on immunologic factors and cell membrane fatty

acids in experimental autoimmune encephalomyelitis. Gene. 2020;759:144987.

71. Hegazy O, Platnick H. Cannabidiol (CBD) for treatment of neurofibromatosis-related pain and concomitant mood disorder: A case report. Cureus. 2019;11(12).

72. Sznitman SR, Vulfsons S, Meiri D, Weinstein G. Medical cannabis and insomnia in older adults with chronic pain: a cross-sectional study. BMJ Supportive & Palliative Care. 2020;10(4):415-20.

73. Erridge S, Miller M, Gall T, Costanzo A, Pacchetti B, Sodergren MH. A comprehensive patient and public involvement program evaluating perception of cannabis-derived medicinal products in the treatment of acute postoperative pain, nausea, and vomiting using a qualitative thematic framework. Cannabis and Cannabinoid Research. 2020;5(1):73-80.

74. Bhatt D, Hazari A, Yamakawa GR, Salberg S, Sgro M, Shultz SR et al. Investigating the cumulative effects of Δ9-tetrahydrocannabinol and repetitive mild traumatic brain injury on adolescent rats. Brain Communications. 2020;2(1):fcaa042.

75. Hegazy O, Platnick H. Cannabidiol (CBD) for treatment of neurofibromatosis-related pain and concomitant mood disorder: A case report. Cureus. 2019;11(12).

76. Magid L, Heymann S, Elgali M, Avram L, Cohen Y, Liraz-Zaltsman S et al. Role of CB2 receptor in the recovery of mice after traumatic brain injury. Journal of neurotrauma. 2019;36(11):1836-46.

77. Aziz MA, Khan AH, Ullah H, Adnan M, Hashem A, Abd_Allah EF. Traditional phytomedicines for gynecological problems used by tribal communities of Mohmand Agency near the Pak-Afghan border area. Revista Brasileira de Farmacognosia. 2018;28:503-11.

78. Singh D, Fisher J, Shagalov D, Varma A, Siegel DM. Dangerous plants in dermatology: Legal and controlled. Clinics in Dermatology. 2018;36(3):399-419.

79. Rein JL, Wyatt CM. Marijuana and cannabinoids in ESRD and earlier stages of CKD. American Journal of Kidney Diseases. 2018;71(2):267-74.

80. Hoffenberg EJ, Newman H, Collins C, Leinwand K, Tarbell S. Cannabis and pediatric inflammatory bowel disease: change blossoms a mile high. Journal of pediatric gastroenterology and nutrition. 2017;64(2):265.

81. Androvicova R, Horacek J, Tintera J, Hlinka J, Rydlo J, Jezova D et al. Individual prolactin reactivity modulates response of nucleus accumbens to erotic stimuli during acute cannabis intoxication: an fMRI pilot study. Psychopharmacology. 2017;234(13):1933-43.

82. Madikizela B, Kambizi L, McGaw LJ. An ethnobotanical survey of plants used traditionally to treat tuberculosis in the eastern region of OR Tambo district, South Africa. South African Journal of Botany. 2017;109:231-6.

83. Fongnzossie EF, Tize Z, Nde PF, Biyegue CN, Ntsama IB, Dibong SD et al. Ethnobotany and pharmacognostic perspective of plant species used as traditional cosmetics and cosmeceuticals among the Gbaya ethnic group in Eastern Cameroon. South African Journal of Botany. 2017;112:29-39.

84. Cheifetz AS, Gianotti R, Luber R, Gibson PR. Complementary and alternative medicines used by patients with inflammatory bowel diseases. Gastroenterology. 2017;152(2):415-29.

85. Kogan NM, Melamed E, Wasserman E, Raphael B, Breuer A, Stok KS et al. Cannabidiol, a major non-psychotropic cannabis constituent enhances fracture healing and stimulates lysyl hydroxylase activity in osteoblasts. Journal of Bone and Mineral Research. 2015;30(10):1905-13.

86. Yeshurun M, Shpilberg O, Herscovici C, Shargian L, Dreyer J, Peck A et al. Cannabidiol for the prevention of graft-versus-host-disease after allogeneic hematopoietic cell transplantation: results of a phase II study. Biology of Blood and Marrow Transplantation. 2015;21(10):1770-5.

87. Mondino A, Cavelli M, González J, Santana N, Castro-Zaballa S, Mechoso B et al. Acute effect of vaporized Cannabis on sleep and electrocortical activity. Pharmacology Biochemistry and Behavior. 2019;179:113-23.