Cancer therapy-The role of cannabinoids and endocannabinoids

Panagiotis Zogopoulos

Department of Neurosurgery, General Hospital of Nikaia-Piraeus “Agios Panteleimon”, Athens, Greece

Correspondence: Panagiotis Zogopoulos
E-mail: p.zogopoulos@yahoo.com
Received: February 01, 2015
Published online: March 22, 2015

Cannabinoids have recently been added to the therapeutic armamentarium of cancer, not only for their palliative effects (inhibition of chemotherapy-induced nausea and vomiting) but also as antitumor drugs, based on their ability to induce cancer cell apoptosis and reduce neoangiogenesis as well as tumor cell invasiveness and metastasis. A large number of researches have reported the antitumor effects of cannabinoids on various cancer types (glioma, breast, prostate, skin, lung, thyroid, gastric, colon, hepatocellular, pancreatic, lymphoma and leukemia). Furthermore, the endogenous signalling system of endocannabinoids which share common receptors and manifest similar actions with cannabinoids, is also a promising field for the treatment of cancer. We present recent advances regarding ongoing research of cannabinoids and endocannabinoids in various cancer types.

Keywords: cancer; cannabinoids; endocannabinoids; glioma; breast; prostate; lung; thyroid; gastrointestinal; leukemia

To cite this article: Panagiotis Zogopoulos. Cancer therapy-The role of cannabinoids and endocannabinoids. Can Cell Microenviorn 2015; 2: e583. doi: 10.14800/ccm.583.

Introduction

Cannabinoids have recently been added to the therapeutic armamentarium of cancer, since they exert palliative effects such as inhibition of chemotherapy-induced nausea and vomiting, appetite stimulation and analgesia. Recent evidence suggests that they may also act as antitumor drugs, based on their ability to induce cancer cell apoptosis and reduce neoangiogenesis as well as tumor cell invasiveness and metastasis. The anti-proliferative effects of cannabinoids have been reported in various cultured cancer cells of neural, breast, prostate, skin and thyroid origin, as well as in lymphoma and leukemia cells [1-8]. Furthermore, the endogenous signalling system of endocannabinoids which share common receptors and manifest similar actions with cannabinoids, is also a promising field for the treatment of cancer. We present recent advances regarding ongoing research of cannabinoids and endocannabinoids in various cancer types.

Cannabinoids and endocannabinoids

Cannabinoids are a class of chemical compounds with numerous pharmacological effects mediated through two specific plasma membrane G-protein-coupled receptors, referred to as cannabinoid receptors (CB) 1 and 2. They include phytocannabinoids (hydrocarbon compounds found in cannabis plant) and other compounds with similar actions or chemical structure to phytocannabinoids [9].

Δ⁹-Tetrahydrocannabinol (Δ⁹-THC) (the primary psychoactive component of cannabis plant), cannabiol (CBN) and cannabidiol (CBD) are the most studied natural cannabinoids [10]. Synthetic cannabinoids are comprised of various chemical classes: the classical cannabinoids with similar structure to Δ⁹-THC, the nonclassical ones (like quinolines, arylysulfonamides, aminoalkylindoles and 1,5-diarylpyrazoles) and eicosanoids related to endocannabinoids [9].
Endocannabinoids, on the other hand, are endogenous metabolites of eicosanoid fatty acids. They are lipid signaling molecules that exert their actions through the same CB receptors with $\Delta^9$-THC$^{[11]}$. They are derivatives of arachidonic acid conjugated with either ethanolamine or glycerol. Apart from anandamide (AEA) and 2-arachidonoylglycerol (2-AG), which are the most studied, other endocannabinoids are 2-arachidonoylglyceryl ether (2-AGE, noladin ether), N-arachidonoyldopamine (NADA) and O-arachidonylethanolamine (OAE, virdohamine)$^{[9]}$.

Cancer types

Over the last years, a growing array of data suggest that cannabinoids, as well as endocannabinoids can be effectively used in the treatment of various cancer types, including gliomas, breast, prostate, skin, lung, thyroid, gastric, colon, hepatocellular and pancreatic cancer, as well as in lymphoma and leukemia. Recent advances are presented for each type separately.

Gliomas

Gliomas are the most common primary brain tumors, accounting for more than 40% of all CNS neoplasms and are highly resistant to available therapeutic approaches, such as radiation and chemotherapy$^{[12,13]}$. Despite recent advances in determining gliomas pathogenesis in a molecular level, and current efforts to develop more effective treatment strategies, these tumors have a generally poor prognosis, with median survival time for patients with advanced tumors (grade III/IV) being approximately 1 year$^{[9,14,15]}$.

Cannabinoids inhibited growth and angiogenesis of gliomas in animal models$^{[16,17]}$. They have been shown to induce apoptosis of glioma cells in vitro and inhibit angiogenesis of gliomas in vivo$^{[5,18]}$. Studies in animal models demonstrated that local administration of $\Delta^9$-THC or the synthetic cannabinoid WIN-55,212-2 (a mixed CB1/CB2 agonist, synthetic aminoalkylindole) reduced the size of tumors generated by intracranial inoculation of C6 glioma cells in rats, without affecting healthy brain tissue. This led to complete glioma eradication and prolonged survival in one third of the cannabinoids-treated rats$^{[19]}$. Studies performed in mouse xenograft models with intratumoral and intraperitoneal drug administration demonstrated that non-psychoactive phytocannabinoid CBD, $\Delta^9$-THC, WIN-55,212-2, JWH133(a CB2-selective agonist) or the novel synthetic cannabinoid KM-233 blocked the proliferation of tumors derived from the rat C6 glioma cell line and also from glioblastoma multiforme (GBM) cells obtained from human tumors implanted subcutaneously in the flank of immune-deficient mice$^{[19-22]}$.

$\Delta^9$-THC administration to mice with human astrocytoma resulted in inhibition of mTOR signaling pathway, increased TRB3 expression, caspase-3 activation and appearance of autophagy markers$^{[23]}$. Such findings indicate that $\Delta^9$-THC promotes autophagy-mediated human glioma cell death through stimulation of endoplasmic reticulum (ER) stress$^{[23]}$. WIN 55,212-2 and $\Delta^9$-THC significantly reduced in vitro the growth of various human GBM cell lines examined, including SF126, U87MG, U251, U373MG, and SF188$^{[24]}$. $\Delta^9$-THC decreases cell proliferation and increases cell death of human GBM cells more rapidly than WIN 55,212-2$^{[24]}$. Cannabinoids may further reduce the invasive potential of cancer cells by inhibiting their adhesion to the vascular endothelium. The adhesion molecules intercellular adhesion molecule-1 (ICAM-1) and vascular cell adhesion molecule-1 (VCAM-1), are a prerequisite for extravasation of circulating cells from blood vessels. They have been shown to be downregulated at the mRNA level after treatment with WIN 55,212-2 in astrocytes$^{[25]}$. These effects, associated to the reduction of VEGF and VEGF receptor-2 (VEGFR-2) levels in glioma cells following cannabinoid administration, might lead to inhibition of angiogenesis in gliomas, thus directly enhancing anti-tumor activity$^{[16,17]}$.

Apart from plant derived (e.g. $\Delta^9$-THC and CBD) and synthetic (e.g. WIN-55,212-2) cannabinoids, endocannabinoids (e.g. AEA and 2-AG) have also been shown to block cancer cell proliferation, induce apoptosis of cancer cells and have neuroprotective effects both in vitro and in vivo, as recently reviewed$^{[18,19,26-29]}$.

Clinical experimental therapies administered intracranially $\Delta^9$-THC via catheter in the resection cavity of glioblastomas and it was found that $\Delta^9$-THC inhibited the facilitated tumor growth in glioblastoma patients$^{[30]}$.

Breast cancer

Breast cancer is the most common malignant disease among Western women and the second leading cause of cancer-related death in women in the United States$^{[31]}$. CBD has been shown to inhibit growth of various breast tumor cell lines (MCF7, MDA-MB-231) in a potent and selective manner, since it exhibited significantly lower potency in non-cancer cells$^{[32,33]}$. Besides proliferation, CBD has also been found to interfere with invasion and metastasization of breast cancer cells$^{[34]}$. CBD regulates the expression of key genes controlling cell proliferation and invasion through the downregulation of Id-1 expression, an inhibitor of basic helix-loop-helix transcription factors$^{[35]}$. Id-1 overexpression in breast cancer cells is responsible for their proliferation, migration and invasion thus, the ability of CBD to
significantly decrease its expression in breast cancer cells might be associated with its efficacy in reducing tumor aggressiveness[35].

Cannabidiolic acid (CBDA), the parent molecule of CBD, inhibited migration of the highly invasive MDA-MB-231 human breast cancer cells in vitro, apparently through a mechanism involving inhibition of cAMP-dependent protein kinase A[36].

Δ9-THC, WIN-55,212-2 and JWH-133, a non-psychotropic CB2 receptor-selective agonist, reduced ErbB2-positive breast cancer tumor growth, tumor number, and the amount/severity of lung metastases in experimental mice[37,38]. Histological analyses of the tumors revealed that cannabinoids inhibit cancer cell proliferation, induce cancer cell apoptosis and impair tumor angiogenesis, at least partially through the inhibition of pro-tumorigenic Akt pathway[38].

Prostate cancer

Prostate cancer is the most common malignancy and one of the leading causes of cancer death among men of all races. In prostate cancer cells, cannabinoids, following receptor binding, inhibit cell proliferation and induce cell cycle arrest and apoptosis through cAMP-PKA/Raf-ERK signalling pathways[39]. CB1 receptor immunoreactivity (CB1IR) in prostate cancer tissues has been associated with disease severity and outcome. Patients with a high tumor CB1IR score had a significantly higher proportion of Gleason scores 8-10, metastases at diagnosis, tumor size and rate of cell proliferation at diagnosis than patients with a lower score, while CB1IR in non-malignant tissue was not associated with disease outcome[40]. Inhibitory actions of AEA on EGFR expression and EGFR-induced proliferation of prostate cancer cells have also been demonstrated, with this effect occurring in a CB1-dependent manner[41].

Δ9-THC, CBD, AEA, 2-AG, 2-AGE, R(+)-Methanandamide, WIN-55,212-2, JWH-015 and HU120 have all exerted anti-proliferative, apoptotic and anti-invasive effects in different prostate cancer cells both in vitro and in vivo[3,4,24,45]. Moreover, WIN55,212-2 decreased androgen receptor (AR) expression and prostate specific antigen (PSA) levels in prostate cancer cells[3,4].

Skin cancer

Cutaneous melanoma is one of the most aggressive human cancers[46]. Metastatic patients have poor prognosis and they are generally refractory to conventional chemotherapy[47]. Cannabinoids have been reported to inhibit in vivo growth of melanomas expressing CB1 and CB2 receptors by decreasing proliferation, angiogenesis and metastasis formation, while increasing apoptosis[46].

AEA, in the micromolar range of concentrations, exerted a cytotoxic effect against human A375 melanoma cells through a complex mechanism, which involves cyclooxygenase (COX)-2 and lipoygenase (LOX)-derived product synthesis and CB1 activation, eventually leading to activation of a caspase-dependent apoptotic pathway[48]. WIN55212-2 induced apoptotic cell death in human melanoma cells which express both CB1 and CB2 receptors. However, none of both CB1 and CB2 receptors, nor vaniloid receptor mediated apoptotic death. Instead, it was suggested that the membrane lipid raft mediates the proapoptotic effects of WIN55212-2 on human melanoma cell lines via the intrinsic caspase pathway[49].

Non-melanoma skin cancer and other epithelial tumors overexpress cyclooxygenase-2 (COX-2), differentiating them from normal cells[50]. In tumorigenic keratinocytes that overexpress COX-2, AEA activated ER stress pathways [PKR-like ER kinase (PERK), inositol requiring kinase-1 (IRE1) and activating transcription factor-6 (ATF6)] and the ER stress apoptosis-associated proteins [C/EBP homologous protein-10 (CHOP10), caspase-12 and caspase-3], thus inducing apoptosis[50]. AEA-stimulated ER stress and apoptosis only occurred in the presence of COX-2, since AEA was metabolized by COX-2 to the novel J-series prostaglandin-ethanolamides (prostamides) that activate apoptotic ER stress[50]. Since endogenous levels of COX-2 are low in non-tumor keratinocytes, AEA may produce selective toxicity in tumor cells and provide an effective approach for topical treatment of non-melanoma skin cancer or other cancers that overexpress COX-2[50].

Lung cancer

Non-small cell lung cancer is one of the leading causes of cancer deaths worldwide[51]. In spite of this fact, only limited therapeutic options are available in the current clinical practice. Although the first anticancer effect of cannabinoids on lung tumors was reported in the 1970s, it was only recently that increased attention was given to their therapeutic potential for this disease[52,53].

A selective CB2 receptor agonist, JWH-133 (3-(1,1-dimethylbutyl)-1-deoxy-Δ8-tetrahydrocannabinol) exerted cytotoxic effect in A549 non-small lung cancer cells in vitro, although at high concentrations (10⁴ mol/l), while inhibition of colony formation was also detected at non-toxic, nano-molar concentrations (10⁻⁵-10⁻⁸ mol/l)[54]. Furthermore, JWH-133 was found to induce weak DNA
fragmentation in A549 cells and also inhibited at non-toxic concentrations (10^4-10^6 mol/l), some steps in the process of angiogenesis (such as endothelial cell migration)\(^{54}\).

CBD has also been shown to inhibit invasiveness of A549 cells, through the activation of both CB receptors, as well as transient receptor potential vanilloid 1 (TRPV1) receptor. This inhibition of A549 cell invasion was also accompanied by the downregulation of another important factor involved in the regulation of cell spreading, the plasminogen activator inhibitor PAI-1\(^{55,56}\). In the lung cancer cell lines A549, H358 and H460, CBD elicited concentration-dependent ICAM-1 up-regulation compared to vehicle via CB receptors, TRPV 1 and p42/44 mitogen activated protein kinase\(^{57}\). Overall, cannabinoids induce ICAM-1, thereby conferring tissue inhibitor of matrix metalloproteinas-1 (TIMP-1) induction and subsequent decreased cancer cell invasiveness\(^{57}\).

**Thyroid cancer**

Thyroid carcinomas are the most frequent malignant endocrine tumors in humans\(^{58}\). Thyroid neoplasias are characterized by tumors with different molecular and clinical features including differentiated follicular and papillary carcinomas, poorly differentiated papillary and follicular carcinomas and undifferentiated anaplastic carcinomas\(^{59-61}\).

2-methyl-2-F-anandamide (Met-F-AEA), a metabolically stable analogue of anandamide, was found effective in inhibiting growth in cell lines derived from thyroid carcinomas\(^{61}\). Growth inhibition was associated with a high level of CB1 receptor expression, suggesting that the cytotoxic effect is due to interaction with the CB1 receptor\(^{61}\). This phenomenon was associated with activation of protein p53 and an increased apoptotic rate\(^{61}\). Furthermore, CB2 agonist JWH-133 and CB1/CB2 agonist WIN-55,212-2 induced apoptosis in anaplastic thyroid carcinoma cells (ARO) and ARO/IL-12 cells\(^{62}\).

**Gastric cancer**

Antitumor activity of cannabinoids on gastric cancers has not been investigated extensively and thus, knowledge regarding mechanisms involved is limited. Treatment of MKN-1 and AGS human gastric cancer cells with WIN 55,212-2 significantly decreased cell survival and induced apoptosis\(^{63,64}\). WIN 55,212-2 treatment activated MAPK pathway and significantly downregulated phosphorylated AKT (pAKT). Inhibition of pAKT led to cell cycle arrest in the G0/G1 phase and eventually to apoptosis in human gastric cancer cells\(^{63,64}\).

In the human gastric cancer cell line, HGC-27, which express CB1 receptor, AEA stimulated proliferation at low concentrations (under 1mM), while it strongly suppressed proliferation through the induction of apoptosis at higher concentrations (10mM)\(^{65}\). When AEA was used with paclitaxel (a chemotherapeutic drug of the taxane family), AEA at 10mM synergistically enhanced the cytotoxic effect of paclitaxel, whereas it showed no significant effect at lower concentrations\(^{65}\). Flow cytometric analysis revealed that addition of 10 mM AEA synergistically enhanced paclitaxel-induced apoptosis, possibly through the activation of caspase-3, -8, and -9\(^{65}\).

**Colon cancer**

Colon cancer is a major cause of morbidity and mortality in Western countries. A recent paper demonstrated the chemopreventive effect of CBD in a preclinical animal model of colon cancer based on azoxymethane (AOM) administration in mice\(^{66}\). AOM treatment was associated with aberrant crypt foci, polyps and tumor formation, as well as with the upregulation of phospho-Akt, iNOS and COX-2 and the downregulation of caspase-3\(^{66}\). CBD was effective in reducing aberrant crypt foci, polyps and tumors and counteracted AOM-induced phospho-Akt and caspase-3 changes\(^{66}\). In in vitro studies with colorectal carcinoma cell lines, CBD protected DNA from oxidative damage, increased endocannabinoid concentrations and reduced cell proliferation, through activation of CB1 and TRPV1 receptors\(^{35,66}\).

CBD receptor has been found to be mainly expressed in human normal colonic epithelium whereas tumor tissue is strongly positive for the CB2 receptor\(^{67}\). Activation of CB1 and, more efficiently, of CB2 receptors induced apoptosis and increased ceramide levels in the DLD-1 and HT29 colon cancer cell lines\(^{67}\). Apoptosis was prevented by the pharmacologic inhibition of ceramide de novo synthesis, therefore it was concluded that either CB1 or CB2 receptor activation induces apoptosis through ceramide de novo synthesis in colon cancer cells\(^{67}\).

**Pancreatic cancer**

Pancreatic cancer is one of the most aggressive and lethal human malignancies. CB1 and CB2 receptors are expressed in both normal and pancreatic cancer tissues \(^{68}\). Cannabinoids can induce alterations of pancreatic cancer cell metabolism (inhibition of glycolytic pathway and Krebs cycle), thus resulting in a strong induction of autophagy and inhibition of cell growth\(^{69}\). They have been demonstrated to inhibit pancreatic cancer cell proliferation both in vitro and in vivo, and autophagy has been demonstrated to mediate this process or to be itself a death mechanism\(^{70}\). Cannabinoid
administration leads to apoptosis of pancreatic tumor cells via CB2 receptor and ceramide-dependent up-regulation of p8 and ATF-4 and TRB3 stress-related genes.[45,71]

**Hepatocellular cancer**

Hepatocellular carcinoma (HCC) is the most frequent primary solid tumor of the liver and it is estimated to account for 5% of all malignant neoplasias[72]. The antitumor activity of cannabinoids against HCC cells has been related to the ability of these drugs to induce apoptosis and autophagy[72]. 

Δ9-THC and JWH-015 have exhibited antitumor effects against HepG2 and HUH-7 HCC cells [72]. Specifically, Δ9-THC and JWH-015 reduced significantly the tumor growth rate in vivo in experimental mice [72]. Pharmacological inhibition of PPARc decreased the cannabinoid-induced cell death and apoptosis, suggesting that PPARc is necessary for the autophagy flux promoted by cannabinoids [72]. Thus, the antiproliferative action of cannabinoids Δ9-THC and JWH-015 on HCC, both in vitro and in vivo, are modulated by upregulation of PPARc-dependent pathways[72].

**Lymphoma and Leukemia**

CBD treatment induced apoptosis, through caspase-3 activation in human acute myeloid leukaemia HL-60 cell line, whereas it had no effect on human monocytes from normal individuals[73]. CBD exposure of murine EL-4 lymphoma cell line and human Jurkat and Molt-4 leukemia cell lines, led to a significant CB2 receptor-mediated decrease in the number of viable cells as well as to the induction of apoptosis, both in vitro and in vivo[74]. Moreover, previous evidence indicated that human leukemias and lymphomas expressed significantly higher levels of CB2 receptors compared with other tumor cell lines, suggesting that tumors of immune origin may be highly sensitive to CB2-mediated effects of CBD[75]. Therefore, CBD acting through CB2 receptors and ROS production, may represent a novel and highly selective treatment for leukaemia and lymphoma.

**Discussion**

Cannabinoids, the active components of marijuana and their other natural and synthetic analogues have been reported as useful adjuvants to conventional chemotherapeutic regimens for preventing nausea, vomiting, pain and for stimulating appetite. Recently, the antitumorigenic effects of cannabinoids have emerged as an exciting field in cancer research and may complement in the future the therapeutic profile of their administration [76,77]. Apart from their proapoptotic and antiproliferative action, recent research has shown that cannabinoids may likewise limit tumor cell angiogenesis, migration, invasion, adhesion and metastasis, by pathways including activation of both cannabinoid receptors as well as TRPV1[78]. The safety of Δ9-THC administration has been determined and a dose escalation regimen showed that cannabinoid delivery was safe and could be achieved without overt psychoactive effects[30].

Besides a great body of experimental evidence pointing to an antitumorigenic action of cannabinoids and endocannabinoids, the lack of severe adverse side effects of these compounds as compared to the generalized toxic actions of conventional chemotherapeutics, makes them promising candidates for the treatment of various cancer types in the future.

**Conclusions**

Precise mechanisms of cannabinoid antitumor actions need to be further investigated with well-designed studies. Since most cannabinoids exhibit a fair safety profile and there is sufficient evidence of their antiproliferative actions on tumor cells, clinical trials are necessary to determine whether cannabinoids could be used for the inhibition of tumor growth in humans. If this could be established, then non-toxic, non-habit-forming cannabinoids could be developed as novel therapeutic agents for the treatment of cancer.

**Conflicting interests**

The author declares that there are no conflicting interests.

**References**


