

Cannabinoids in Gynecological Diseases

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Keywords

Endocannabinoid system · Cannabinoid receptors · Endometriosis · Cervical cancer · TRPV1

Abstract

The endocannabinoid system (ECS) is a multifunctional homeostatic system involved in many physiological and pathological conditions. The ligands of the ECS are the endocannabinoids, whose actions are mimicked by exogenous cannabinoids, such as phytocannabinoids and synthetic cannabinoids. Responses to the ligands of the ECS are mediated by numerous receptors like the classical cannabinoid receptors (CB₁ and CB₂) as well as ECS-related receptors, e.g., G protein-coupled receptors 18 and 55 (GPR18 and GPR55), transient receptor potential ion channels, and nuclear peroxisome proliferator-activated receptors. The ECS regulates almost all levels of female reproduction, starting with oocyte production through to parturition. Dysregulation of the ECS is associated with the development of gynecological disorders from fertility disorders to cancer. Cannabinoids that act at the ECS as specific agonists or antagonists may potentially influence dysregulation and, therefore, represent new therapeutic options for the therapy of gynecological disorders.

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Cannabinoids and the Endocannabinoid System

Cannabinoids: Endo-, Phyto-, and Synthetic Cannabinoids

Since its first description as a multifunctional system 2 decades ago, the endocannabinoid system (ECS) has gained a lot of interest [1]. The ECS comprises enzymes, cannabinoid receptors and their related receptors, and ligands, i.e., the endocannabinoids (eCB), which are synthesized endogenously. Phytocannabinoids (pCB) that are isolated from *Cannabis sativa* and synthetic cannabinoids (sCB) affect the receptors of the ECS as exogenous cannabinoids.

The first eCB that were discovered were N-arachidonoyl-ethanolamine, better known as anandamide (AEA), and 2-arachidonoylglycerol (2-AG) [2–4]. Further endogenous ligands of the ECS are 2-AG ether (noladin ether), N-arachidonoyl dopamine, and O-arachidonoyl ethanolamine (virodhamine) [5–7]. The best investigated eCB are AEA and 2-AG, which are produced “on demand”. They are triggered by a stimulus that leads to an increase in the intracellular Ca²⁺ concentration and cleavage of precursor molecules [6, 7]. Synthesis of eCB take place in several tissues and cell types where they are

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catalyzed by specific synthases, such as N-acylphosphatidylethanolamine-hydrolyzing phospholipase D (NAPE-PLD) and others [8]. After release, inactivation of AEA and 2-AG occurs promptly by enzymatic hydrolysis of the amide and ester bonds by fatty acid amide hydrolase (FAAH) and monoacylglycerol lipase (MAGL) [9–13].

More than a 100 pCB have been identified, of which the psychotropic (–)-*trans*- Δ^9 -tetrahydrocannabinol (THC) and the nonpsychotropic (–)-cannabinol (CBD) are the best studied [14–16]. THC and CBD mediate a broad spectrum of biological actions including analgesic, antiemetic, and anti-inflammatory effects [12–14].

Classical Cannabinoid Receptors

Responses to eCB, but also to pCB and sCB, are mediated by numerous receptors of which cannabinoid receptor type 1 (CB₁) and cannabinoid receptor type 2 (CB₂) represent the classical cannabinoid receptors. CB₁ and CB₂ are G-protein-coupled receptors and they are involved in many (patho-) physiological processes such as pain, inflammation, cancer, and hypertension as well as neurodegenerative disorders [15]. The expression of the CB₁ receptor in the brain is responsible for the psychotropic effects of THC and other synthetic CB₁ agonists. CB₁ expression has also been found in peripheral organs like the heart, spleen, and endocrine glands as well as in parts of the male and female reproductive systems and the urinary tract, including the ovaries, uterus, testis, prostate, and placenta [16–19]. The second classical cannabinoid receptor, CB₂, is expressed only to a minor degree in the nervous system. It is mainly located in tissues of the immune system including the spleen, tonsils, thymus, and bone marrow as well as in immune cells such as B cells, natural killer cells, monocytes, neutrophils, and CD8⁺ and CD4⁺ T cells [20–22].

ECS-Related Receptors

Besides CB₁ and CB₂, many other cannabinoid-sensitive receptors exist that can be designated as ECS-related receptors. Several studies have emphasized their relationship with the ECS [23–25]. The metabotropic G-protein-coupled receptors GPR55, GPR18, and GPR119 have been demonstrated to be targets of eCB, but also of pCB and sCB, but they have not been categorized as cannabinoid receptors by the International Union of Pharmacology [25–29]. Other families of ECS-related receptors are the nuclear peroxisome proliferator-activated receptors (PPAR) and the transient receptor potential (TRP) ion channels [30–33]. The GPR55 receptor plays an important role in cancer cell behavior. It has been shown by dif-

ferent groups in the last few years that GPR55 is involved in cancer cell proliferation in vitro and/or in vivo in various types of cancers including ovarian, prostate, and skin cancer as well as non-small lung cancer [34–38].

PPAR represent a family of nuclear hormone receptors consisting of 3 isoforms (α , δ , and γ [39]), and they are expressed in many organs including the ovaries, uterus, and prostate [40–42]. Numerous functions have been attributed to these receptors including the regulation of metabolism and energy homeostasis, cell proliferation, and inflammation [43–45]. These effects are mediated by a multitude of endogenous and exogenous ligands, e.g., eicosanoids or plant extracts [46, 47]. Within the last 2 decades, researchers have shown that cannabinoids (i.e., eCB, pCB, and sCB) mediate anti-inflammatory and antiemetic effects also via PPAR α and PPAR γ receptors [48–53].

The TRP channel superfamily responds to many physical and chemical stimuli, including cannabinoids [54]. TRP channels that cause proliferative effects belong to the 3 major subfamilies of these channels, i.e., the TRPC (canonical), the TRPV (vanilloid), and the TRPM (melastatin) channels [55]. Within the members of these subfamilies the TRPV6 channel is the best studied. A high expression of TRPV6 has been found in many types of cancers, such as colon, thyroid, prostate, and ovarian cancer [56–61]. The TRPC6 channel of the TRPC subfamily and some members of the TRPM subfamily have been shown to be related to procarcinogenic effects in prostate, cervical, ovarian, breast, and gastric cancers [62–67].

(Patho-) Physiological Impact of the ECS

A variety of physiological and pathological processes throughout the organism are affected by the ECS including modulation of neuronal functions, microcirculation, and functions of immune cells [68]. Hence, the ECS takes part in the modulation of pain and inflammation and may be also involved in regulatory processes during carcinogenesis [69–75]. Ligands of the ECS could act via cannabinoid receptors as well as via ECS-related receptors. The receptors represent therapeutic opportunities in the treatment of pain, inflammation, and chemotherapy-induced nausea or vomiting since they cause inhibitory effects in these pathological processes [76]. Aside from that, modulation of the ECS by natural and synthetic ligands may also result in the induction of apoptosis, inhibition of cancer cell invasion, and neoangiogenesis [77–81].

The ECS in Gynecological Disorders and Cancer

Since the ECS is involved in almost all levels of female reproduction, i.e., from oocyte production to parturition, several studies in recent years have shown that dysregulation of the ECS is associated with the development of disorders of the female reproductive tract [82–90]. These include fertility disorders like polycystic ovary syndrome (PCOS), endometriosis, and gynecological cancers [91–93].

Cannabinoids and PCOS

PCOS is a metabolic and endocrinal disorder. Its pathogenesis was only recently connected to the ECS by demonstrating that levels of AEA and 2-AG were elevated in peripheral blood mononuclear cells of women with PCOS [94]. Recently, Cui et al. [95, 96] found reduced FAAH expression in the endometrium of patients with PCOS and an increase in AEA plasma levels, as FAAH is mainly involved in AEA degradation. Previous studies have shown that elevated plasma AEA levels in connection with a lower FAAH activity resulted in ectopic pregnancy which is also linked to PCOS [85, 97]. Thus, it seems likely that a dysregulation of the ECS is involved in pregnancy complications of women with PCOS.

Apart from high eCB levels, insulin resistance is common in the pathophysiology of PCOS, often causing hepatic stress and liver damage, which ends up in elevated levels of alanine aminotransferase, a marker of hepatocellular injury. Dawson et al. [91] recently reported that a weight-reducing therapy with the CB₁ antagonist rimonabant in obese women with PCOS resulted in a reduction of alanine aminotransferase, accompanied by a reduction of insulin resistance. In contrast, it has been reported that the amount of both vascular endothelial growth factor and interleukin-8, which play a crucial role in inflammation, are paradoxically increased upon rimonabant treatment of obese women with PCOS, which may compensate the benefit associated with weight loss [98]. More data are, therefore, needed to clarify the impact of the ECS in the pathogenesis of PCOS and whether a CB₁ antagonist may be of benefit.

Cannabinoids and Endometriosis

Endometriosis is a disease characterized by ectopic growth of uterine endometrial tissue and it is usually associated with severe pain. Since the mechanisms for endometriosis-related pain can be divided into 3 categories (nociceptive, inflammatory, and neuropathic), each of which is linked to the ECS, it is not surprising that the

ECS represents a big field of research for the development of new therapeutic tools in the management of this disorder. Sanchez et al. [99] recently reported elevated plasma levels of AEA and 2-AG in women with endometriosis. There were, however, no changes in CB₁ expression in endometrial stromal cells during the menstrual cycle of the women with endometriosis, although in the healthy controls (and in contrast to findings by Bilgic et al. [100]) an upregulation of CB₁ was found in the S-phase [99]. TRPV1, an ECS-related receptor, was found to be expressed at comparable levels in ectopic endometrial stromal cells from both healthy controls and women with endometriosis [99]. These findings are in accordance with previously published studies showing TRPV1 expression in ectopic endometrial epithelial cells [101]. The presence of this receptor in endometrial tissue and the elevated levels of eCB in patients with endometriosis may therefore be associated with the development of chronic inflammatory pain [99].

Besides the discovery of new treatment targets for pain management, there is still the need to know how ectopic lesions develop and proliferate. By immunostaining of CB₁ and CB₂ in endometriotic and normal tissue, Bilgic et al. [100] showed that the expression of both receptors was reduced in the glandular epithelial and stromal cells of women with endometriosis. This is in agreement with findings by Resuehr et al. [88], who observed reduced immunostaining of CB₁ in patients with endometriosis [88, 100]. Moreover, it was demonstrated that the selective CB₁ agonist ACPA as well as the selective CB₂ agonist CB 65 induced apoptosis and reduced proliferation of Ishikawa cells (normal endometrial glandular cells) and the endometriosis cyst wall cells CRL-7566 [100]. The CB₂-mediated effect was more prominent in Ishikawa cells while the CB₁-mediated effect was more prominent in CLR-7566 cells [100]. Similar results were presented earlier by Leconte et al. [102], who demonstrated an antiproliferative effect of the nonselective CB₁/CB₂ agonist WIN 55212-2 on deep infiltrating endometrial stromal cells. This effect was a result of inactivation of the Akt pathway by WIN 55212-2 [102]. These *in vitro* results were confirmed by a mouse model of deep infiltrating endometriosis [102]. In contrast, Sanchez et al. [103] found that selective activation of CB₁ by methanandamide was linked to the development of ectopic lesions in a mouse model of endometriosis. The discrepancies may be explained by the fact that there are basic differences in the animal models of endometriosis. Sanchez et al. [103] used a model to elucidate the development of ectopic lesions in initial stages while Leconte et al. [102] transplanted human endometriotic tissue into nude

mice to investigate effects on already established lesions. Thus, species differences between the rodent and human endometrium may exist [102, 103]. Certainly, more research is necessary to identify potential new targets for therapy of endometriosis.

Cannabinoids and Cervical Cancer

Cervical cancer is the second leading cause of cancer in women and, due to the lack of effective treatment, more than 250,000 deaths are reported annually [104]. A possible influence of the ECS in the development of cervical cancer has been elucidated in recent years. Contassot et al. [89] reported a strong expression pattern of CB₁ and CB₂ as well as TRPV1 in cervical carcinoma cell lines and biopsies. On top of that, it was shown that AEA had proapoptotic effects on cervical carcinoma cell lines (HeLa and Caski) [89], which were not inhibited but rather enhanced by CB₁ and CB₂ antagonists. On the other hand, the TRPV1 selective antagonist capsazepine protected the cell lines from AEA-induced apoptosis, indicating an important role of the TRPV1 channel in the proapoptotic action of AEA [89]. Additionally, it was demonstrated by Ramer et al. [105] that CBD is able to decrease the invasiveness of cancer cells in a concentration-dependent manner. The effect was observed in the cervical cancer cell lines HeLa and C33A as well as in the lung cancer cell line A549 and seemed to be mediated by the upregulation of TIMP-1 via CB₁/CB₂ and TRPV1. The activation of p38 and p42/44 mitogen-activated protein kinases was identified as an upstream event of TIMP-1 upregulation [105]. In agreement with these findings, it was reported that treatment of different cervical cancer cell lines (HeLs, SiHa, ME-180) with CBD led to a decrease of cell proliferation [106]. Furthermore, CBD induced cell death by the accumulation of cells in the sub-G₀ phase (cell death phase) of the cell cycle, a finding that was most likely caspase dependent because caspase-9 as well as caspase-3 were upregulated upon CBD treatment [106]. Hence, CBD may be an additional therapeutic tool for the treatment of cervical cancer, but *in vivo* studies will be needed to exactly clarify the impact of CBD on cervical cancer.

Cannabinoids and Ovarian Cancer

Among gynecological cancers, ovarian cancer is responsible for the highest mortality rate [107]. To determine a possible role of the ECS in the pathophysiology of the ovaries, El-Talatini et al. [82] studied the expression levels of different components of the ECS [108]. They were able to show the expression of CB₁ and CB₂ as well as of NAPE-PLD and FAAH in normal human ovaries

by immunohistochemical methods. Additionally, they found AEA in the follicular fluid after ovarian stimulation by hormones (following an *in vitro* fertilization protocol which caused an increase in follicle size), suggesting that AEA is involved in the maturation of follicles and oocytes [82, 108]. Another study group demonstrated expression of CB₁ and FAAH in the ovarian surface epithelium from which ovarian cancers originate, which could be another hint for a possible involvement of the ECS in ovarian cancer [109]. The 2-AG degrading enzyme MAGL has been shown to be upregulated in aggressive human ovary cancer cells [110]. MAGL seems to be involved in oncogenic signaling and hence in increased migration, invasion, and survival of cancer cells. This was also demonstrated by MAGL overexpression in nonaggressive cancer cells which subsequently exhibited an increased pathogenic phenotype [110]. Moreover, the application of an MAGL inhibitor led to a reversion of the enhanced pathogenicity [110]. Regarding the expression of CB₁ in ovarian cancer, Messalli et al. [111] revealed, by using immunohistochemistry, that CB₁ expression was moderate in benign and borderline epithelial ovarian tumors, but the expression was strongly increased in invasive ovarian tumors. These findings suggest a correlation between the expression patterns of ECS components and the prognosis for ovarian cancer malignancy [111].

It also turned out that the amount of lysophospholipids in blood and ascites fluids was elevated in ovarian cancer patients compared to healthy controls, a finding associated with proliferation and the metastatic potential of ovarian cancer cells [112]. Hofman et al. [38] more recently described that the elevation of lysophosphatidylinositol (an endogenous GPR55 agonist) in the ovarian cancer cell lines OVCAR-3, OVCAR-5, and COV-362 resulted in a GPR55-dependent angiogenesis because pharmacological inhibition and genetic deletion of GPR55 reduced the proangiogenic potential of lysophosphatidylinositol in these cell lines. Additionally, they demonstrated that the mitogen-activated protein kinase pathway was activated via GPR55 by phosphorylation of ERK1/2 and p38, which are signaling molecules known to be involved in proliferative and migratory responses [38]. Thus, the involvement of the ECS in ovarian cancer may fuel expectations on new therapeutics to combat this type of cancer.

Cannabinoids and Endometrial Cancer

Endometrial cancer can be classified into type I and II tumors. Depending on the disease stage, various therapies exist, but the prognosis is still poor because of tumor recurrence [113]. Guida et al. [114] reported an upregula-

tion of CB₂ in endometrial cancer, whereby immunostaining was only successful in transformed malignant cells and completely absent in normal endometrial tissue. Furthermore, 2-AG levels were increased but MAGL expression was decreased in comparison to controls, while AEA levels and FAAH expression were unaffected [114]. In accordance with these findings, Jové et al. [115] demonstrated that CB₂ was expressed at higher levels in stages III and IV of endometrial carcinoma, which has been linked to a poor prognosis. Contrary to Guida et al. [114], they found an increase in CB₁ expression by immunohistochemistry in endometrial carcinoma tissue compared to normal endometrial tissue [115].

The effects of pCB, such as THC, on endometrial cancer progression was recently evaluated by Zhang et al. [116]. They found that THC inhibited endometrial cancer cell proliferation and migration by a decreased expression of matrix metalloproteinase-9 but not matrix metalloproteinase-2. The same effects could be detected after matrix metalloproteinase-9 silencing [116]. More recently, the involvement of THC and CBD in endometrial cancer was investigated in 2 model cell lines, i.e., Ishikawa and Hec50co cells [117]. Expression of all components of the ECS, including TRPV1, was detected in the cells. Additionally, treatment of the cells with AEA or CBD (>5 µM) resulted in a reduced cell viability that was linked to an increase in ROS production and caspase-3/-7 activity [117]. The findings regarding the proapoptotic action of AEA are well in accordance with the observations by Contassot et al. [89], who described AEA-driven cancer cell apoptosis via TRPV1 activation.

References

- Zuardi AW. Cannabidiol: from an inactive cannabinoid to a drug with wide spectrum of action. *Br J Psychiatry*. 2008 Sep;30(3):271–80.
- Devane WA, Hanus L, Breuer A, Pertwee RG, Stevenson LA, Griffin G, et al. Isolation and structure of a brain constituent that binds to the cannabinoid receptor. *Science*. 1992 Dec; 258(5090):1946–9.
- Mechoulam R, Ben-Shabat S, Hanus L, Ligumsky M, Kaminski NE, Schatz AR, et al. Identification of an endogenous 2-monoglyceride, present in canine gut, that binds to cannabinoid receptors. *Biochem Pharmacol*. 1995 Jun;50(1):83–90.
- Sugiura T, Kondo S, Sukagawa A, Nakane S, Shinoda A, Itoh K, et al. 2-Arachidonoylglycerol: a possible endogenous cannabinoid receptor ligand in brain. *Biochem Biophys Res Commun*. 1995 Oct;215(1):89–97.
- Porter AC, Sauer JM, Knierman MD, Becker GW, Berna MJ, Bao J, et al. Characterization of a novel endocannabinoid, virodhamine, with antagonist activity at the CB1 receptor. *J Pharmacol Exp Ther*. 2002 Jun;301(3):1020–4.
- Matias I, Di Marzo V. Endocannabinoid synthesis and degradation, and their regulation in the framework of energy balance. *J Endocrinol Invest*. 2006;29(3 Suppl):15–26.
- Giuffrida A, Beltramo M, Piomelli D. Mechanisms of endocannabinoid inactivation: biochemistry and pharmacology. *J Pharmacol Exp Ther*. 2001 Jul;298(1):7–14.
- Maccarrone M, Guzmán M, Mackie K, Doherty P, Harkany T. Programming of neural cells by (endo)cannabinoids: from physiological rules to emerging therapies. *Nat Rev Neurosci*. 2014 Dec;15(12):786–801.
- Karlsson M, Contreras JA, Hellman U, Tornqvist H, Holm C. cDNA cloning, tissue distribution, and identification of the catalytic triad of monoglyceride lipase. Evolutionary relationship to esterases, lysophospholipases, and haloperoxidases. *J Biol Chem*. 1997 Oct; 272(43):27218–23.
- Dinh TP, Carpenter D, Leslie FM, Freund TF, Katona I, Sensi SL, et al. Brain monoglyceride lipase participating in endocannabinoid inactivation. *Proc Natl Acad Sci USA*. 2002 Aug; 99(16):10819–24.
- Oláh A, Szekanecz Z, Bíró T. Targeting cannabinoid signaling in the immune system: "high"-ly exciting questions, possibilities, and challenges. *Front Immunol*. 2017 Nov;8:1487.
- Bueb JL, Lambert DM, Tschirhart EJ. Receptor-independent effects of natural cannabinoids in rat peritoneal mast cells in vitro. *Biochim Biophys Acta*. 2001 Apr;1538(2-3):252–9.

Conclusions

A pivotal effect of the ECS in gynecological disorders and cancers was demonstrated by various working groups in recent years. In particular, the development, progression and prognosis of female urogenital diseases seem to be associated with dysregulation of the ECS. Due to its regulatory functions, the ECS represents an important therapeutic target to be elucidated. Cannabinoids, especially pCB or sCB, that manipulate the ECS as specific agonists or antagonists may potentially influence dysregulation. For this reason, more research is required to shed light on the complex interactions of the ECS in order to find new therapeutic tools for therapy of gynecological disorders and cancer.

Acknowledgement

R.S. is supported by the Austrian Science Fund (FWF grants P30144 and KLI521-B31).

Statement of Ethics

The authors have no ethical conflicts to disclose.

Disclosure Statement

The authors have no conflict of interests to declare.

- 13 Ligresti A, De Petrocellis L, Di Marzo V. From Phytocannabinoids to Cannabinoid Receptors and Endocannabinoids: Pleiotropic Physiological and Pathological Roles Through Complex Pharmacology. *Physiol Rev*. 2016 Oct;96(4):1593–659.
- 14 Solymosi K, Köfalvi A. Cannabis: a treasure trove or Pandora's box? *Mini Rev Med Chem*. 2017;17(13):1223–91.
- 15 Pertwee RG. Targeting the endocannabinoid system with cannabinoid receptor agonists: pharmacological strategies and therapeutic possibilities. *Philos Trans R Soc Lond B Biol Sci*. 2012 Dec;367(1607):3353–63.
- 16 Pertwee RG. Pharmacology of cannabinoid CB1 and CB2 receptors. *Pharmacol Ther*. 1997;74(2):129–80.
- 17 Taylor AH, Ang C, Bell SC, Konje JC. The role of the endocannabinoid system in gametogenesis, implantation and early pregnancy. *Hum Reprod Update*. 2007 Sep-Oct;13(5):501–13.
- 18 Galiègue S, Mary S, Marchand J, Dussosoy D, Carrière D, Carayon P, et al. Expression of central and peripheral cannabinoid receptors in human immune tissues and leukocyte subpopulations. *Eur J Biochem*. 1995 Aug;232(1):54–61.
- 19 Kenney SP, Kekuda R, Prasad PD, Leibach FH, Devoe LD, Ganapathy V. Cannabinoid receptors and their role in the regulation of the serotonin transporter in human placenta. *Am J Obstet Gynecol*. 1999 Aug;181(2):491–7.
- 20 Parolaro D, Massi P, Rubino T, Monti E. Endocannabinoids in the immune system and cancer. *Prostaglandins Leukot Essent Fatty Acids*. 2002 Feb-Mar;66(2-3):319–32.
- 21 Pertwee RG. Sites and Mechanisms of Action. In: Grotenhermen F, Russo E, editors. Cannabis and cannabinoids: pharmacology, toxicology, and therapeutic potential. Binghamton: Haworth Press; 2002. p. 73–88.
- 22 Habayeb OM, Taylor AH, Bell SC, Taylor DJ, Konje JC. Expression of the endocannabinoid system in human first trimester placenta and its role in trophoblast proliferation. *Endocrinology*. 2008 Oct;149(10):5052–60.
- 23 Morales P, Hurst DP, Reggio PH. Molecular targets of the phytocannabinoids: a complex picture. In: Kinghorn AD, Gibbons S, editors. *Phytocannabinoids*. Cham: Springer; 2017. p. 103–31.
- 24 Pertwee RG, Howlett AC, Abood ME, Alexander SP, Di Marzo V, Elphick MR, et al. International Union of Basic and Clinical Pharmacology. LXXIX. Cannabinoid receptors and their ligands: beyond CB₁ and CB₂. *Pharmacol Rev*. 2010 Dec;62(4):588–631.
- 25 Stephen A [Internet]. IUPHAR/BPS Guide to Pharmacology: GPR18, GPR55, and GPR119. Available from: <http://www.guidetopharmacology.org/GRAC/FamilyDisplayForward?familyId=114>.
- 26 Pertwee RG, Abood M, Alexander SPH, et al. [Internet]: IUPHAR/BPS Guide to Pharmacology: Cannabinoid Receptors. Available from: <http://www.guidetopharmacology.org/GRAC/FamilyDisplayForward?familyId=13>.
- 27 Ross RA. The enigmatic pharmacology of GPR55. *Trends Pharmacol Sci*. 2009 Mar;30(3):156–63.
- 28 Qin Y, Verdegaal EM, Siderius M, Bebelman JP, Smit MJ, Leurs R, et al. Quantitative expression profiling of G-protein-coupled receptors (GPCRs) in metastatic melanoma: the constitutively active orphan GPCR GPR18 as novel drug target. *Pigment Cell Melanoma Res*. 2011 Feb;24(1):207–18.
- 29 Okuno T, Yokomizo T. What is the natural ligand of GPR55? *J Biochem*. 2011 May;149(5):495–7.
- 30 Godlewski G, Offertáler L, Wagner JA, Kunos G. Receptors for acylethanolamides-GPR55 and GPR119. *Prostaglandins Other Lipid Mediat*. 2009 Sep;89(3-4):105–11.
- 31 Caterina MJ. TRP channel cannabinoid receptors in skin sensation, homeostasis, and inflammation. *ACS Chem Neurosci*. 2014 Nov;5(11):1107–16.
- 32 O'Sullivan SE. An update on PPAR activation by cannabinoids. *Br J Pharmacol*. 2016 Jun;173(12):1899–910.
- 33 Laprairie RB, Bagher AM, Denovan-Wright EM. Cannabinoid receptor ligand bias: implications in the central nervous system. *Curr Opin Pharmacol*. 2017 Feb;32:32–43.
- 34 Piñeiro R, Maffucci T, Falasca M, Píeiro R, Maffucci T, Falasca M, et al. The putative cannabinoid receptor GPR55 defines a novel autocrine loop in cancer cell proliferation. *Oncogene*. 2011 Jan;30(2):142–52.
- 35 Pérez-Gómez E, Andradas C, Flores JM, Quintanilla M, Paramio JM, Guzmán M, et al. The orphan receptor GPR55 drives skin carcinogenesis and is upregulated in human squamous cell carcinomas. *Oncogene*. 2013 May;32(20):2534–42.
- 36 Adinolfi B, Romanini A, Vanni A, Martinotti E, Chicca A, Fogli S, et al. Anticancer activity of anandamide in human cutaneous melanoma cells. *Eur J Pharmacol*. 2013 Oct;718(1-3):154–9.
- 37 He D, Wang J, Zhang C, Shan B, Deng X, Li B, et al. Down-regulation of miR-675-5p contributes to tumor progression and development by targeting pro-tumorigenic GPR55 in non-small cell lung cancer. *Mol Cancer*. 2015 Apr;14(1):73.
- 38 Hofmann NA, Yang J, Trauger SA, Nakayama H, Huang L, Strunk D, et al. The GPR 55 agonist, L- α -lysophosphatidylinositol, mediates ovarian carcinoma cell-induced angiogenesis. *Br J Pharmacol*. 2015 Aug;172(16):4107–18.
- 39 Alexander SP, Cidlowski JA, Kelly E, Marrion N, Peters JA, Benson HE, et al; CGTP Collaborators. The Concise Guide to PHARMACOLOGY 2015/16: nuclear hormone receptors. *Br J Pharmacol*. 2015 Dec;172(24):5956–78.
- 40 Komar CM, Braissant O, Wahli W, Curry TE Jr. Expression and localization of PPARs in the rat ovary during follicular development and the periovarial period. *Endocrinology*. 2001 Nov;142(11):4831–8.
- 41 Froment P, Fabre S, Dupont J, Pisselet C, Chesneau D, Staels B, et al. Expression and functional role of peroxisome proliferator-activated receptor-gamma in ovarian folliculogenesis in the sheep. *Biol Reprod*. 2003 Nov;69(5):1665–74.
- 42 Mouihate A, Boissé L, Pittman QJ. A novel antipyretic action of 15-deoxy-Delta^{12,14}-prostaglandin J₂ in the rat brain. *J Neurosci*. 2004 Feb;24(6):1312–8.
- 43 Friedland SN, Leong A, Filion KB, Genest J, Lega IC, Mottillo S, et al. The cardiovascular effects of peroxisome proliferator-activated receptor agonists. *Am J Med*. 2012 Feb;125(2):126–33.
- 44 Menendez-Gutierrez MP, Roszer T, Ricote M. Biology and therapeutic applications of peroxisome proliferator-activated receptors. *Curr Top Med Chem*. 2012;12(6):548–84.
- 45 Debril MB, Renaud JP, Fajas L, Auwerx J. The pleiotropic functions of peroxisome proliferator-activated receptor γ . *J Mol Med (Berl)*. 2001;79(1):30–47.
- 46 Kliewer SA, Sundseth SS, Jones SA, Brown PJ, Wisely GB, Koble CS, et al. Fatty acids and eicosanoids regulate gene expression through direct interactions with peroxisome proliferator-activated receptors alpha and gamma. *Proc Natl Acad Sci USA*. 1997 Apr;94(9):4318–23.
- 47 Wang L, Waltenberger B, Pferschy-Wenzig EM, Blunder M, Liu X, Malainer C, et al. Natural product agonists of peroxisome proliferator-activated receptor gamma (PPAR γ): a review. *Biochem Pharmacol*. 2014 Nov;92(1):73–89.
- 48 O'Sullivan SE, Kendall DA, Randall MD. Time-dependent vascular effects of Endocannabinoids mediated by peroxisome proliferator-activated receptor gamma (PPAR γ). *PPAR Res*. 2009;2009:425289.
- 49 O'Sullivan SE. Cannabinoids go nuclear: evidence for activation of peroxisome proliferator-activated receptors. *Br J Pharmacol*. 2007 Nov;152(5):576–82.
- 50 O'Sullivan SE, Tarling EJ, Bennett AJ, Kendall DA, Randall MD. Novel time-dependent vascular actions of Δ^9 -tetrahydrocannabinol mediated by peroxisome proliferator-activated receptor gamma. *Biochem Biophys Res Commun*. 2005 Nov;337(3):824–31.
- 51 Sun Y, Alexander SP, Garle MJ, Gibson CL, Hewitt K, Murphy SP, et al. Cannabinoid activation of PPAR alpha; a novel neuroprotective mechanism. *Br J Pharmacol*. 2007 Nov;152(5):734–43.
- 52 Granja AG, Carrillo-Salinas F, Pagani A, Gómez-Cañas M, Negri R, Navarrete C, et al. A cannabigerol quinone alleviates neuroinflammation in a chronic model of multiple sclerosis. *J Neuroimmune Pharmacol*. 2012 Dec;7(4):1002–16.

- 53 Bouaboula M, Hilairat S, Marchand J, Fajas L, Le Fur G, Casellas P. Anandamide induced PPARgamma transcriptional activation and PP13-L1 preadipocyte differentiation. *Eur J Pharmacol*. 2005 Jul;517(3):174–81.
- 54 Di Marzo V, De Petrocellis L. Endocannabinoids as regulators of transient receptor potential (TRP) channels: A further opportunity to develop new endocannabinoid-based therapeutic drugs. *Curr Med Chem*. 2010;17(14):1430–49.
- 55 Shapovalov G, Ritaine A, Skryma R, Prevarskaya N. Role of TRP ion channels in cancer and tumorigenesis. *Semin Immunopathol*. 2016 May;38(3):357–69.
- 56 Bolanz KA, Hediger MA, Landowski CP. The role of TRPV6 in breast carcinogenesis. *Mol Cancer Ther*. 2008 Feb;7(2):271–9.
- 57 Bödding M, Fecher-Trost C, Flockerzi V. Store-operated Ca²⁺ current and TRPV6 channels in lymph node prostate cancer cells. *J Biol Chem*. 2003 Dec;278(51):50872–9.
- 58 Peng JB, Zhuang L, Berger UV, Adam RM, Williams BJ, Brown EM, et al. CaT1 expression correlates with tumor grade in prostate cancer. *Biochem Biophys Res Commun*. 2001 Apr;282(3):729–34.
- 59 Vanden Abeele F, Roudbaraki M, Shuba Y, Skryma R, Prevarskaya N. Store-operated Ca²⁺ current in prostate cancer epithelial cells. Role of endogenous Ca²⁺ transporter type 1. *J Biol Chem*. 2003 Apr;278(17):15381–9.
- 60 Wissenbach U, Niemeyer B, Himmerkus N, Fixemer T, Bonkhoff H, Flockerzi V. TRPV6 and prostate cancer: cancer growth beyond the prostate correlates with increased TRPV6 Ca²⁺ channel expression. *Biochem Biophys Res Commun*. 2004 Oct;322(4):1359–63.
- 61 Zhuang L, Peng JB, Tou L, Takana H, Adam RM, Hediger MA, et al. Calcium-selective ion channel, CaT1, is apically localized in gastrointestinal tract epithelia and is aberrantly expressed in human malignancies. *Lab Invest*. 2002 Dec;82(12):1755–64.
- 62 Singh J, Manickam P, Shmoish M, Natik S, Denyer G, Handelsman D, et al. Annotation of androgen dependence to human prostate cancer-associated genes by microarray analysis of mouse prostate. *Cancer Lett*. 2006 Jun;237(2):298–304.
- 63 Schinke EN, Bii V, Nalla A, Rae DT, Tedrick L, Meadows GG, et al. A novel approach to identify driver genes involved in androgen-independent prostate cancer. *Mol Cancer*. 2014 May;13(1):120.
- 64 Armisen R, Marcelain K, Simon F, Tapia JC, Toro J, Quest AF, et al. TRPM4 enhances cell proliferation through up-regulation of the β -catenin signaling pathway. *J Cell Physiol*. 2011 Jan;226(1):103–9.
- 65 Kiessling A, Füssel S, Schmitz M, Stevanovic S, Meye A, Weigle B, et al. Identification of an HLA-A*0201-restricted T-cell epitope derived from the prostate cancer-associated protein trp-p8. *Prostate*. 2003 Sep;56(4):270–9.
- 66 Hopkins MM, Feng X, Liu M, Parker LP, Koh DW. Inhibition of the transient receptor potential melastatin-2 channel causes increased DNA damage and decreased proliferation in breast adenocarcinoma cells. *Int J Oncol*. 2015 May;46(5):2267–76.
- 67 Kahl CR, Means AR. Regulation of cell cycle progression by calcium/calmodulin-dependent pathways. *Endocr Rev*. 2003 Dec;24(6):719–36.
- 68 Maurya N, Velmurugan BK. Therapeutic applications of cannabinoids. *Chem Biol Interact*. 2018 Sep;293:77–88.
- 69 Fine PG, Rosenfeld MJ. The endocannabinoid system, cannabinoids, and pain. *Rambam Maimonides Med J*. 2013 Oct;4(4):e0022.
- 70 Disis ML. Immune regulation of cancer. *J Clin Oncol*. 2010 Oct;28(29):4531–8.
- 71 Pisanti S, Borselli C, Oliviero O, Laezza C, Gazzero P, Bifulco M. Antiangiogenic activity of the endocannabinoid anandamide: correlation to its tumor-suppressor efficacy. *J Cell Physiol*. 2007 May;211(2):495–503.
- 72 Pisanti S, Picardi P, D'Alessandro A, Laezza C, Bifulco M, et al. The endocannabinoid signaling system in cancer. *Trends Pharmacol Sci*. 2013 May;34(5):273–82.
- 73 De Petrocellis L, Orlando P, Moriello AS, Avilello G, Stott C, Izzo AA, et al. Cannabinoid actions at TRPV channels: effects on TRPV3 and TRPV4 and their potential relevance to gastrointestinal inflammation. *Acta Physiol (Oxf)*. 2012 Feb;204(2):255–66.
- 74 Borrelli F, Fasolino I, Romano B, Capasso R, Maiello F, Coppola D, et al. Beneficial effect of the non-psychotropic plant cannabinoid cannabigerol on experimental inflammatory bowel disease. *Biochem Pharmacol*. 2013 May;85(9):1306–16.
- 75 Liu YJ, Fan HB, Jin Y, Ren CG, Jia XE, Wang L, et al. Cannabinoid receptor 2 suppresses leukocyte inflammatory migration by modulating the JNK/c-Jun/Alox5 pathway. *J Biol Chem*. 2013 May;288(19):13551–62.
- 76 Grant I, Atkinson JH, Gouaux B, Wilsey B. Medical marijuana: clearing away the smoke. *Open Neurol J*. 2012;6(1):18–25.
- 77 Athanasiou A, Clarke AB, Turner AE, Kumaran NM, Vakilpour S, Smith PA, et al. Cannabinoid receptor agonists are mitochondrial inhibitors: a unified hypothesis of how cannabinoids modulate mitochondrial function and induce cell death. *Biochem Biophys Res Commun*. 2007 Dec;364(1):131–7.
- 78 Maccarrone M, Lorenzon T, Bari M, Melino G, Finazzi-Agro A. Anandamide induces apoptosis in human cells via vanilloid receptors. Evidence for a protective role of cannabinoid receptors. *J Biol Chem*. 2000 Oct;275(41):31938–45.
- 79 Kim SR, Lee DY, Chung ES, Oh UT, Kim SU, Jin BK. Transient receptor potential vanilloid subtype 1 mediates cell death of mesencephalic dopaminergic neurons in vivo and in vitro. *J Neurosci*. 2005 Jan;25(3):662–71.
- 80 Curran S, Murray GI. Matrix metalloproteinases: molecular aspects of their roles in tumor invasion and metastasis. *Eur J Cancer*. 2000 Aug;36(13 Spec No):1621–30.
- 81 Freimuth N, Ramer R, Hinz B. Antitumorigenic effects of cannabinoids beyond apoptosis. *J Pharmacol Exp Ther*. 2010 Feb;332(2):336–44.
- 82 El-Talatini MR, Taylor AH, Elson JC, Brown L, Davidson AC, Konje JC. Localisation and function of the endocannabinoid system in the human ovary. *PLoS One*. 2009;4(2):e4579.
- 83 Schuel H, Burkman LJ, Lippes J, Crickard K, Forester E, Piomelli D, et al. N-acyl ethanolamines in human reproductive fluids. *Chem Phys Lipids*. 2002 Dec;121:211–27.
- 84 Wang H, Guo Y, Wang D, Kingsley PJ, Marnett LJ, Das SK, et al. Aberrant cannabinoid signaling impairs oviductal transport of embryos. *Nat Med*. 2004 Oct;10(10):1074–80.
- 85 Gebeh AK, Willets JM, Marczylo EL, Taylor AH, Konje JC. Ectopic pregnancy is associated with high anandamide levels and aberrant expression of FAAH and CB1 in fallopian tubes. *J Clin Endocrinol Metab*. 2012 Aug;97(8):2827–35.
- 86 Taylor AH, Abbas MS, Habiba MA, Konje JC. Histomorphometric evaluation of cannabinoid receptor and anandamide modulating enzyme expression in the human endometrium through the menstrual cycle. *Histochem Cell Biol*. 2010 May;133(5):557–65.
- 87 Fonseca BM, Correia-da-Silva G, Taylor AH, Lam PM, Marczylo TH, Konje JC, et al. N-acyl ethanolamine levels and expression of their metabolizing enzymes during pregnancy. *Endocrinology*. 2010 Aug;151(8):3965–74.
- 88 Resuehr D, Glore DR, Taylor HS, Bruner-Tran KL, Osteen KG. Progesterone-dependent regulation of endometrial cannabinoid receptor type 1 (CB1-R) expression is disrupted in women with endometriosis and in isolated stromal cells exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD). *Fertil Steril*. 2012 Oct;98(4):948–56.e1.
- 89 Contassot E, Tenan M, Schnüriger V, Pelte MF, Dietrich PY. Arachidonyl ethanolamide induces apoptosis of uterine cervix cancer cells via aberrantly expressed vanilloid receptor-1. *Gynecol Oncol*. 2004 Apr;93(1):182–8.
- 90 Eichele K, Ramer R, Hinz B. R(+)-methanandamide-induced apoptosis of human cervical carcinoma cells involves a cyclooxygenase-2-dependent pathway. *Pharm Res*. 2009 Feb;26(2):346–55.
- 91 Dawson AJ, Kilpatrick ES, Coady AM, Elshewehy AM, Dakroury Y, Ahmed L, et al. Endocannabinoid receptor blockade reduces alanine aminotransferase in polycystic ovary syndrome independent of weight loss. *BMC Endocr Disord*. 2017 Jul;17(1):41.
- 92 Pertwee RG. Emerging strategies for exploiting cannabinoid receptor agonists as medicines. *Br J Pharmacol*. 2009 Feb;156(3):397–411.

- 93 Ayakannu T, Taylor AH, Willets JM, Konje JC. The evolving role of the endocannabinoid system in gynaecological cancer. *Hum Reprod Update*. 2015 Jul-Aug;21(4):517–35.
- 94 Juan CC, Chen KH, Wang PH, Hwang JL, Seow KM. Endocannabinoid system activation may be associated with insulin resistance in women with polycystic ovary syndrome. *Fertil Steril*. 2015 Jul;104(1):200–6.
- 95 Cui N, Feng X, Zhao Z, Zhang J, Xu Y, Wang L, et al. Restored Plasma Anandamide and Endometrial Expression of Fatty Acid Amide Hydrolase in Women With Polycystic Ovary Syndrome by the Combination Use of Diane-35 and Metformin. *Clin Ther*. 2017 Apr;39(4):751–8.
- 96 Cui N, Yang Y, Xu Y, Zhang J, Jiang L, Hao G. Decreased expression of fatty acid amide hydrolase in women with polycystic ovary syndrome. *Gynecol Endocrinol*. 2017 May;33(5):368–72.
- 97 Gebeh AK, Willets JM, Bari M, Hirst RA, Marczylo TH, Taylor AH, et al. Elevated anandamide and related N-acyl ethanolamine levels occur in the peripheral blood of women with ectopic pregnancy and are mirrored by changes in peripheral fatty acid amide hydrolase activity. *J Clin Endocrinol Metab*. 2013 Mar;98(3):1226–34.
- 98 Sathyapalan T, Javed Z, Kilpatrick ES, Coady AM, Atkin SL. Endocannabinoid receptor blockade increases vascular endothelial growth factor and inflammatory markers in obese women with polycystic ovary syndrome. *Clin Endocrinol (Oxf)*. 2017 Mar;86(3):384–7.
- 99 Sanchez AM, Cioffi R, Viganò P, Candiani M, Verde R, Piscitelli F, et al. Elevated Systemic Levels of Endocannabinoids and Related Mediators Across the Menstrual Cycle in Women With Endometriosis. *Reprod Sci*. 2016 Aug;23(8):1071–9.
- 100 Bilgic E, Guzel E, Kose S, Aydin MC, Karaismailoglu E, Akar I, et al. Endocannabinoids modulate apoptosis in endometriosis and adenomyosis. *Acta Histochem*. 2017 Jun;119(5):523–32.
- 101 Rocha MG, e Silva JC, Ribeiro da Silva A, Candido Dos Reis FJ, Nogueira AA, Polineto OB. TRPV1 expression on peritoneal endometriosis foci is associated with chronic pelvic pain. *Reprod Sci*. 2011 Jun;18(6):511–5.
- 102 Leconte M, Nicco C, Ngô C, Arkwright S, Chéreau C, Guibourdenche J, et al. Antiproliferative effects of cannabinoid agonists on deep infiltrating endometriosis. *Am J Pathol*. 2010 Dec;177(6):2963–70.
- 103 Sanchez AM, Quattrone F, Pannese M, Ulisse A, Candiani M, Diaz-Alonso J, et al. The cannabinoid receptor CB1 contributes to the development of ectopic lesions in a mouse model of endometriosis. *Hum Reprod*. 2017 Jan;32(1):175–84.
- 104 International Agency for Research on Cancer [Internet]. GLOBOCAN 2012 v1.0 (2013): Cancer Incidence and Mortality Worldwide – IARC CancerBase No. 11. Available from: [https://www.scirp.org/\(S\(351jmbntvnsjt1aadkposzje\)\)/reference/ReferencesPapers.aspx?ReferenceID=1817155](https://www.scirp.org/(S(351jmbntvnsjt1aadkposzje))/reference/ReferencesPapers.aspx?ReferenceID=1817155).
- 105 Ramer R, Merkord J, Rohde H, Hinz B. Cannabidiol inhibits cancer cell invasion via up-regulation of tissue inhibitor of matrix metalloproteinases-1. *Biochem Pharmacol*. 2010 Apr;79(7):955–66.
- 106 Lukhele ST, Motadi LR. Cannabidiol rather than Cannabis sativa extracts inhibit cell growth and induce apoptosis in cervical cancer cells. *BMC Complement Altern Med*. 2016 Sep;16(1):335.
- 107 Siegel RL, Miller KD, Jemal A. Cancer statistics, 2018. *CA Cancer J Clin*. 2018 Jan;68(1):7–30.
- 108 El-Talatini MR, Taylor AH, Konje JC. The relationship between plasma levels of the endocannabinoid, anandamide, sex steroids, and gonadotrophins during the menstrual cycle. *Fertil Steril*. 2010 Apr;93(6):1989–96.
- 109 Bagavandoss P, Grimshaw S. Temporal and spatial distribution of the cannabinoid receptors (CB1, CB2) and fatty acid amide hydrolase in the rat ovary. *Anat Rec (Hoboken)*. 2010 Aug;293(8):1425–32.
- 110 Nomura DK, Long JZ, Niessen S, Hoover HS, Ng SW, Cravatt BF. Monoacylglycerol lipase regulates a fatty acid network that promotes cancer pathogenesis. *Cell*. 2010 Jan;140(1):49–61.
- 111 Messalli EM, Grauso F, Luise R, Angelini A, Rossiello R. Cannabinoid receptor type 1 immunoreactivity and disease severity in human epithelial ovarian tumors. *Am J Obstet Gynecol*. 2014 Sep;211(3):234.e1–6.
- 112 Xu Y, Xiao YJ, Baudhuin LM, Schwartz BM. The role and clinical applications of bioactive lysolipids in ovarian cancer. *J Soc Gynecol Investig*. 2001 Jan-Feb;8(1):1–13.
- 113 Bokhman JV. Two pathogenetic types of endometrial carcinoma. *Gynecol Oncol*. 1983 Feb;15(1):10–7.
- 114 Guida M, Ligresti A, De Filippis D, D'Amico A, Petrosino S, Cipriano M, et al. The levels of the endocannabinoid receptor CB2 and its ligand 2-arachidonoylglycerol are elevated in endometrial carcinoma. *Endocrinology*. 2010 Mar;151(3):921–8.
- 115 Jové M, Gatus S, Yeramian A, Portero-Otin M, Eritja N, Santacana M, et al. Metabotyping human endometrioid endometrial adenocarcinoma reveals an implication of endocannabinoid metabolism. *Oncotarget*. 2016 Aug;7(32):52364–74.
- 116 Zhang Y, Zheng W, Shen K, Shen W. Δ9-tetrahydrocannabinol inhibits epithelial-mesenchymal transition and metastasis by targeting matrix metalloproteinase-9 in endometrial cancer. *Oncol Lett*. 2018 Jun;15(6):8527–35.
- 117 Fonseca BM, Correia-da-Silva G, Teixeira NA. Cannabinoid-induced cell death in endometrial cancer cells: involvement of TRPV1 receptors in apoptosis. *J Physiol Biochem*. 2018 May;74(2):261–72.