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A review on the role of the endocannabinoid system in the gynecological malignancy

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ABSTRACT

Objectives: The endocannabinoid system (ECS) is made up of an array of endogenous bioactive lipids, their receptors and enzymes for their synthesis and degradation. The main endogenous ligands are unsaturated fatty acid derivatives such as anandamide (AEA) and 2-arachidonoylglycerol (2-AG), but many others are still under study. Endocannabinoids are involved in both physiological and pathological conditions and could play an important role in the regulation of processes which lead to cancer.

Methods: With focus on gynaecological cancers, main papers and review articles, up to September 2018, on the role of the ECS, were acquired by PubMed searches using the search terms: 'cannabinoid', 'endocannabinoid', 'gynaecology', 'cancer', and 'malignancy'.

Results: The present review showed the involvement of the endocannabinoid system in numerous physiological and pathological conditions of the female genital tract up to the development of gynaecological malignancy as cervical, endometrial and ovarian cancer.

Conclusion: The endocannabinoid system has an important role in antitumor actions involving different signalling receptor and receptor-independent pathways. It represents an exciting challenge to researchers for its potential use in diagnosis and treatment of all gynaecological malignancies.

Keywords: endocannabinoid system; gynaecological cancers; endometrial cancer; cervical cancer; ovarian cancer; CB1R; CB2R; FAAH

SOMMARIO

Scopo: Il sistema endocannabinoide (ECS) è costituito da una serie di lipidi bioattivi endogeni, i loro recettori e gli enzimi per la loro sintesi e degradazione. I principali ligandi endogeni sono derivati di acidi grassi insaturi come anandamide (AEA) e 2-arachidonoilglicerolo (2-AG), ma molti altri sono ancora in fase di studio. Gli endocannabinoidi sono coinvolti in condizioni fisiologiche e patologiche e potrebbero svolgere un ruolo importante nella regolazione della cancerogenesi.

Materiali e metodi: sono stati acquisiti tramite motore di ricerca PubMed i principali articoli e gli articoli di revisione, fino a dicembre 2018, sul ruolo dell'ECS nei tumori ginecologici, utilizzando i termini di ricerca: "cannabinoide", "endocannabinoide", "ginecologia", "cancro" "e" malignità ".

Risultati: La presente revisione ha mostrato il coinvolgimento del sistema endocannabinoide in numerose condizioni fisiologiche e patologiche del tratto genitale femminile fino allo sviluppo della neoplasia ginecologica come cancro cervicale, endometriale e ovarico.

Conclusioni: Il sistema endocannabinoide ha un ruolo importante nell'azione antitumorali che coinvolge diversi pathways. Rappresenta una sfida entusiasmante per i ricercatori per il suo potenziale utilizzo nella diagnosi e nel trattamento di tutte le neoplasie ginecologiche

INTRODUCTION

The endocannabinoid system (ECS) is made up of an array of endogenous bioactive lipids, their receptors and enzymes for their synthesis and degradation[(1)]. The main endogenous ligands are unsaturated fatty acid derivatives such as anandamide (AEA), 2-arachidonoylglycerol(2-AG), 2-arachidonoylglycerol ether, o-arachidonoyl-ethanolamine, and N-arachidonoyl-dopamine (NADA)[(2,3,4,5)]. These substances are synthetized in various tissues and immune cells by specific synthetases such as N-acylphosphatidylethanolamine-hydrolyzing phospholipase D (NAPE-PLD) for AEA and its congeners and diacylglycerol lipase (DAGL) for

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2-AG[4]. The main enzyme involved in degradation of these substances is the fatty acid amine hydrolase (FAAH) which degrades AEA in arachidonic acid and ethanolamine[6]. Therefore NAPE-PLD and FAAH are the main regulators of the concentration of AEA in target tissues and they are responsible of the so-called anandamide tone[7]. The synthesis of endocannabinoids, in particular AEA and 2-AG, is triggered by the increase of intracellular calcium level[8] and their precursors are phospholipids of cellular membrane. As for receptors, there are two G-protein coupled receptors involved in this system of signaling, CB1R and CB2R. CB1R is the main receptor in the central nervous[9] system but it is also represented in peripheral tissues such as adrenal gland, ovaries, uterus, testis, prostate[10] and placenta[11]. In recent years, CB1R and, more generally, the ECS are isolated and studied in the gastrointestinal tract[12]. The ECS seems to play an important role in the gastrointestinal motility, secretion and sensitivity and to be involved in the regulation of intestinal inflammation and mucosal barrier permeability, which cause functional and pathological disorders[12-18]. CB2R is instead less represented than CB1R in the central nervous system, except during extreme stress conditions[19,20], whereas it has been found in immune-based tissues[21] such as spleen, tonsils, thymus, bone marrow, B-cells, natural killer cells, monocytes, polymorphic mononuclear cells, neutrophils and T8- and T4-postive cells and in the 1st trimester trophoblast[22]. Recent studies highlighted other cannabinoid putative receptors and in particular three G-protein coupled receptors have been related to the ECS. They are GPR55, GPR119[23,24] and GPR18[25]. GPR55 has been identified in human reproductive tract and its ligands are three endocannabinoids, 2-arachidonoylglycerol (2-AG)[26], N-palmitoylethanolamide (PEA) and 2-arachidonolyl lysophosphatidyl-inositol[23]. GPR55 is involved in the development of human squamous skin cancer as its concentration increase in this pathology[27]. Another receptor involved in biological effects of endocannabinoids is a ligand-gated, Ca2+ permeable ion channel, the transient potential vanilloid receptor 1 (TRPV1)[25] that has a ubiquitous distribution[28]. The ligands of this receptor are N-Arachidonoylethanolamide, anandamide (AEA), and N-arachidonoyldopamine (NADA), therefore they can be included in a class of molecules called endovanilloides[28]. Other potential receptors are the peroxisome proliferator-activated receptors (PPARs) which are activated by endocannabinoids in physiological and pathological situations[29]. Endocannabinoids are involved in many biological processes such as metabolic syndrome[30], chronic inflammation including inflammatory bowel disease[31-33], immunomodulation[34], various neurophysiological effects[21, 35] and cancer related pain[36, 37]. ECS could play an important role in the regulation of processes which lead to cancer[38-42] and there is an interesting relation with sex steroid hormone-related cancers[43, 44]. One of the important effects of ECS is the regulation of cellular death. In response to environmental stimuli, the activation of CB1R regulates cell proliferation, differentiation and death thanks to the modulation of the balance among ERK, JNK and p38 MAPK activities[45]. Endocannabinoids can induce apoptosis by activation of TRPVI receptor. Through this interaction, AEA mediates cellular death in rat and human neurons [46, 47], lymphoma cells[47] and cytotrophoblasts[48]. Ligands of cannabinoid receptors generally induce apoptosis inhibiting mitochondrial activity because they increase mitochondrial hydrogen peroxide production and decrease the consumption of oxygen and the mitochondrial membrane potential[49]. Another important effect of cannabinoid receptor activation is the inhibition of cancer cells invasion. These cells use metalloproteinases (MMP-9 and MMP-2) to degrade major basement membrane components, such as laminin, collagen and nidogens[50]. The activation of cannabinoid receptors indirectly down regulates the expression and the activity of MMP-2 through stimulation of TIMP-I which is an inhibitor of the metalloproteinase itself [51]. Endocannabinoids are also thought to be involved in the inhibition of neoangiogenesis decreasing the production of pro-angiogenic factors and/or by directly modulating endothelial cells[52] and decreasing the expression of VEGF and VEGF-R[53,54].

MATERIALS AND METHODS

Literature data up to august 2018 on ECS role in gynecological cancer were acquired in PubMed using the following key words: 'cannabinoid', 'endocannabinoid', 'cancer', 'gynaecology, 'malignancy'. Non-English articles were excluded.

RESULTS

The Endocannabinoid system and gynaecologic Cancer

The recent literature data showed the involvement of the endocannabinoid system in numerous physiological and pathological processes of the female genital tract[55-64]. It is not surprising that a deregulation of the ECS might be involved in the development of gynaecological malignancy[65-67].

The endocannabinoid System ad Endometrial Cancer

Different studies confirmed the presence of some components of the ECS in the uterus and have assessed their function. AEA has been related to decidualization, implantation and menstruation[64, 68-71]. CB1R is mostly present in glandular epithelium and its expression increases during the secretory phase[61, 62]. CB2R is expressed in glands and stroma[61], NAPE-PLD is expressed in the early proliferative phase of menstrual cycle[61] and FAAH is mainly expressed in stroma[61]. Regarding ECS, the most important marker for endometrial carcinoma is CB2R. In the western blot and immunohistochemical analysis, CB2R is mostly expressed in endometrial cancer biopsies conversely it is weakly expressed in normal endometrial tissue[72]. Other studies confirmed also that AEA levels are increased in cancer tissues[72, 73]. CB2R probably plays a key role in the regulation of growth and death of cancer cells[74] and its pathway is altered in endometrial cancer[72]. In this type of cancer, the levels of CB2R and 2-AG are probably increased due to the imbalance of estrogen/progesterone ratio[72]. A recent study on a cellular line of endometrial cancer (AN3CA) showed that CB2R has a potential role in the control of cancer cells growth through the regulation of mitochondrial function and their apoptosis[72]. In the last few years a new kind of research called metabolomics[75] has been used in cancer research. This discipline examines the change of metabolic processes in cancer cells and compares the metabolism of these cells with normal cells of the same tissues to understand physiopathologic processes involved in cancer progression and to find new biomarkers potentially useful to discover new therapies[76, 77]. Furthermore, metabolomics studies demonstrated that there are differences in cellular metabolism between myometrial invasion front and non-invasive endometrial cancer cells[78]. In fact, about invasive front there is an increased expression of endocannabinoid-like metabolites like stearamide and, although the features of stearamide are yet to be demonstrated, this could suppose the potential involvement of ECS in tumor invasion[78]. A recent study showed that, maybe, the endocannabinoids are acting as antagonist of the estrogen and, they could contrast increasing level of sex hormones playing a protective role in the development of hormones related cancers[79]. Another study analyzed the effect of endocannabinoids and phytocannabinoids (cannabidiol, CBD), on cell viability in two endometrial cancers cell lines (Ishikawa cells and Hec50co). This study made clear that ECS and cannabidiol negatively influenced cancer cells viability at concentration higher than 5 μM by increasing levels of caspase -3/-7, cleaved PARP and reactive oxygen species generation. These data suggested apoptosis as the main mechanism involved. This effect is obtained through the stimulation of TRPV1 that leads to an increase of intracellular calcium level[80]. According to all these considerations it is clear that ECS plays an active role in physiopathology of endometrial cancer and it is mandatory to go ahead with scientific research in order to find new potential pharmacological target useful in the care of this cancer.

The Endocannabinoid System and Cervical Cancer

RT-PCR and Western blot analyses have confirmed the presence of CB1, CB2 and TRPV1 receptors in different cervical cancer cell lines [81]. The effect of AEA in cervical cancer has been examined in three cellular lines, which are Caski, HeLa and CC299. AEA determines apoptosis of cancer cells through a mechanism that is not mediated by CB1R and CB2R because their selective antagonists have demonstrated a synergic effect with AEA [79]. In fact, the addition of selective antagonists (SR141716A and SR144528) to CB1 and/or CB2, increased the toxic effects of AEA, suggesting that both CB1 and CB2 are able to protect Cervical Cancer cells from AEA[81]. On the other hand, the selective antagonist of TRPVIR (capsazepine) has been observed to have a protective effect against the action of AEA. For this reason, CB1R and CB2R are protecting cervical cancer cells against the effect of AEA and on the contrary the activation of TRPVI is involved with AEA in cellular death [81]. Another molecule involved in the stimulation of apoptosis in cervical cancer cells is Methanandamide (MA) that is a FAAH-resistant endocannabinoid analogue. MA stimulates apoptosis through the activation of COX-2 by a ceramide-dependent pathway[82]. Furthermore, MA is involved in the inhibition of cancer cell's invasion with a time and concentration dependent mechanism which determines the increase of the expression of TIMP-I[83].

The Endocannabinoid System and Ovarian Cancer

Immunohistochemical examination of CB1R, CB2R, Fatty Acid Amide Hydrolase (FAAH) and N-acyclphosphatidylethanolamine-phospholipase D (NAPE-PLD) in normal human ovaries showed a wide expression of the endocannabinoid system in the ovarian medulla and cortex. Furthermore, evident data suggested that anandamide is produced in the ovary and it is under hormonal control playing a role in folliculogenesis, preovulatory follicle maturation, oocyte maturation and ovulation[55, 84]. The demonstration of CB1R and FAAH expression primarily in the Ovarian Surface Epithelium (OSE) was intriguing. The historical belief that OSE

was the main source of ovarian cancer led to the assessment of the ECS expression in this tumor[85]. Functional proteomic analysis[86] of (aggressive and non-aggressive) ovarian cancer samples proved that aggressive cancer cells show highly increased monoacylglycerol hydrolytic activity and most of this originates from the Monoacylglycerol lipase (MAGL) enzyme[67], which degrades 2-AG. MAGL is strongly expressed in aggressive human ovarian cancer cells, where it modulates a fatty acid network enriched in oncogenic signaling lipids that promotes invasion, migration, survival, and in vivo tumor growth. Overexpression of MAGL in non-aggressive cancer cells recapitulates this fatty acid network and increases their pathogenic phenotypes that are reversed by an MAGL inhibitor[67]. Recently, the orphan receptor G protein-coupled receptor 55 (GPR55) has been suggested as a potential cannabinoid receptor. Elevated GPR55 expression was shown in several ovarian cancer cell lines having a critical role in regulating proliferation. GPR55 mediates the effects of the endogenous ligand lysophosphatidylinositol (LPI), which activates Akt, calcium mobilization and extracellular signal-regulated kinase (ERK) 1/2 expression. Moreover, its down-regulation using small interfering RNA and pharmacological blockade strongly inhibits cell proliferation with important implication as a potential therapeutic target[87].

Ovarian carcinoma cell-derived LPI stimulated angiogenesis in the chorioallantoic membrane (CAM) assa[88]. Applied LPI stimulates proliferation, network formation and migration of neonatal endothelial colony-forming cells (ECFCs) in vitro and angiogenesis in the vivo[88]. The pharmacological GPR55 inhibitor restrained LPI-stimulated ECFC proliferation, network formation and migration in vitro as well as ovarian carcinoma cell- and LPI-induced angiogenesis in vivo[88]. Lately, a variable expression of the endocannabinoid system in human epithelial ovarian tumors was shown. The recorded data demonstrated that the expression of CB1R increased from benign and borderline to malignant ovarian tumors[66]. All the recorded data provide indication that endocannabinoid system could be used in clinical practice to identify or better characterize ovarian tumors, without considering the great opportunity that they might represent as therapeutic targets.

DISCUSSION

The management of women affected by gynecological cancers needs a particular attention even to the preservation of an appropriate quality of life and sexual function, which risk to be severely impaired by surgical/chemo/radio treatments[89-92]. Different lines of evidence support the possibility to use specific biomarkers to identify early stage gynaecological cancers and, in this way, offer a better prognosis to the patients[93-96]. Available data demonstrate that the endocannabinoid system may have a decisive role in the development and progression of gynaecological malignancy. Several mechanisms have been involved in the antitumor actions and include cytotoxic or cytostatic effects, apoptosis induction, and anti-metastatic effects such as the inhibition of neoangiogenesis, tumor cell migration, invasion, and adhesion. The endocannabinoid system activity is very complex, involving different signaling receptors and receptor-independent pathways. For this reason, further studies are needed to understand the exact role of the ECS and its complex activity. There is no doubt that the ECS represents an exciting challenge to researchers for its potential use in diagnosis and treatment of all gynaecological malignancies.

COMPETING INTERESTS

The authors declare neither conflict of interest nor financial and non-financial competing interests

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