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REVIEW ARTICLE

Joint problems arising from lack of repair mechanisms: can cannabinoids help?

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Osteoarthritis OA) is the most common disease of joints, which are complex organs where cartilage, bone and synovium cooperate to allow a range of movements. During progression of the disease, the function of all three main components is jeopardized. Nevertheless, the involvement of each tissue in OA development is still not established and is the topic of the present review. The OA therapies available are symptomatic, largely targeting pain management rather than disease progression. The strong need to develop a treatment for cartilage degeneration, bone deformation and synovial in ammation has led to research on the involvement of the endocannabinoid system in the development of OA. The current review discusses the research on this topic to date and notes the advantages of exploiting endocannabinoid system modulation for cartilage, bone and synovium homeostasis, which could prevent the further progression of OA.

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Abbreviations

2-AG, 2-aracydonylglycerol; AEA, anandamide; CB_1 receptor, cannabinoid receptor 1; CBD, cannabidiol; CCL2, chemokine C-C motif) ligand 2; FAAH, fatty acid amide hydrolase; FLS, broblast-like synoviocyte; GPR18/55, GPCR18/55; OA, osteoarthritis; RA, rheumatoid arthritis; RANK, receptor activator for NF B; RANKL, RANK ligand; TRPV1, transient receptor potential vanilloid 1

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Introduction

Osteoarthritis OA) is the most common type of arthritis, and is a painful degenerative joint disease. It usually affects larger joints that are responsible for weight bearing and is thus one of the main causes of motor disability worldwide. The occurrence of OA increases with age and is more common in women than in men Aigner *et al*., 2004). The joint is an organ constituted mainly of cartilage, synovial broblasts and bone tissue, and although OA was traditionally thought to be a cartilage-based disease, it is now known that during its development, all three components cartilage, subchondral bone and synovial broblasts) may be affected and can contribute to disease progression. OA was previously classi ed as a non-inflammatory type of arthritis. Nevertheless, recent reports indicate that some inflammation occurs and that pro-inflammatory cytokines are released into the joint Robinson *et al*., 2016). Furthermore, a group of OA patients has reported neuropathic pain-like symptoms, which may be a result of damage to the nerves underlying cartilage in the joint Sofat and Kuttapitiya, 2014). OA has a complex origin and various phenotypes, so we would like to discuss the involvement of cartilage, subchondral bone and synovium in the context of OA.

Most of the currently used treatment paradigms in OA are focused on alleviating pain, which should counteract the reduction in a patient s mobility Sarzi-Puttini *et al*., 2005). Nonsteroidal anti-inflammatory drugs, which are often administered to OA patients, can lead to adverse reactions of the gastrointestinal system. Intraarticular injections of corticosteroids may only temporarily alleviate joint pain, and intraarticular injections of hyaluronic acid only bene t some patients, and the effect wears off over time Priano, 2017).

The development of disease-modifying drugs for OA is still in the primary phase. Cytokines are treatment targets, and their inhibition could reduce the inflammatory component of OA Lubberts *et al*., 1998; McDougall *et al*., 2017*;* Wei *et al*., 2017). Inhibitors of metalloproteinases, which degrade the extracellular matrix, have shown potential in preclinical studies, but clinical trials in humans were unsatisfactory Milner and Cawston, 2005). The lack of success in disease-modifying therapies of OA may lie in the timing of the intervention and poor regeneration abilities of joint tissues. Perhaps the highest potential is in preventing the disease onset and treatment at early stages rather than after severe pain symptoms occur.

One promising target for OA-focused therapy is the endocannabinoid system. Cannabinoids are a family of lipid mediators that act through the GPCRs: **CB_1** and **CB_2**, which were originally identi ed as classical cannabinoid receptors Matsuda *et al*., 1990*;* Munro *et al*., 1993) and are the main focus of this review. Several exogenous cannabinoids have been isolated from the *Cannabis* plant, where they occurred naturally; however, most of them are a result of chemical synthesis. Phytocannabinoids are characterized as an agonist of both cannabinoid receptors, whereas synthetic compounds have been produced to act selectively on one of the receptors. In contrast to exogenous ligands, both CB_1 and CB_2 receptors respond to endogenous ligands, such as **anandamide** AEA) and **2-aracydonylglycerol** 2-AG). The endocannabinoid system is a promising target in many different therapies because it is mainly switched on following injury or in the presence of a disease Russo, 2016). Moreover, it is noteworthy that cannabinoid receptors can be manipulated directly by their ligands or indirectly by compounds that modulate levels of endocannabinoids e.g. inhibitors of their degradation enzymes that result in the accumulation of AEA or 2-AG). Cannabinoid ligands in particular AEA) are known to activate other targets, such as **transient receptor potential vanilloid type 1** TRPV1), **GPCR55** GPR55) and **GPR18** GPR18).

The endocannabinoid system has been recognized as playing an important role in the contribution to OA disease progression and has gained increasing interest in recent years. Furthermore, cannabinoids have been proven to possess anti-inflammatory properties and even reduce joint damage during OA Dunn *et al*., 2014). Activation of the endocannabinoid system has been shown to modulate inflammation. The endocannabinoid system is postulated to modulate macrophage and mast cell activation, pro-inflammatory cytokines and T-cell proliferation and apoptosis Robinson *et al*., 2016).

CB_1 receptors are located mainly in the central and peripheral nervous system, while CB_2 receptors are predominantly expressed by immune cells both macrophages in the periphery and glial cells in theCNS) Mbvundula *et al*., 2006). Moreover, both cannabinoid receptors have been shown to be expressed in chondrocyte cultures and human OA cartilage Dunn *et al*., 2014), subchondral bone Rossi *et al*., 2009*;* Whyte *et al*., 2012) and synovial tissue Richardson *et al*., 2008). Synovial broblasts also express AEA and 2-AG catabolic enzymes, suggesting the endocannabinoid system has a role in the maintenance of joint homeostasis McPartland, 2008). Moreover, Schuelert *et al*. showed that CB_1 and CB_2 receptors are expressed on nerve ends that innervate the knee Schuelert and McDougall, 2009). All this evidence suggests that the endocannabinoid system may play an important role in development of joint diseases associated with chronic pain as in OA.

Effects of cannabinoids on osteoarthritic cartilage

In the joint, a thin layer of articular cartilage covers the bone surfaces, providing a durable and pressure-resistant buffer zone that prevents damage in subchondral bone. Cartilage is composed of articular chondrocytes surrounded by an extracellular matrix that contains type II collagen, which is responsible for resistance, and large sulfated proteoglycans, which have a negative charge that enables them to embed water within the tissue to adapt when weight is loaded onto the joint Lories and Luyten, 2012).

The main feature of OA is the loss of articular cartilage and degradation of cartilage matrix, which is primarily attributed to cartilage breakdown. The fundamental role of cartilage in the progression of OA has been well established Goldring, 2000b). However, chondrocytes isolated from OA patients are very diverse, ranging from normal healthy) cells to those with severe changes in both morphology and metabolism, which must be considered in human tissue studies. Chondrocytes maintain cartilage homeostasis and regulate

the stability between anabolic and catabolic processes Krustev *et al.*, 2015). Cartilage degeneration can arise from ageing, trauma, low-grade local or systemic inflammation, metabolic syndromes, obesity and genetic predispositions Buckwalter and Mankin, 1998). The main process contributing to this degeneration is the proteolysis of proteoglycans and collagens, which are major structural components of the cartilage matrix. Pro-inflammatory cytokines promote the breakdown of type II collagen and proteoglycans in the cartilage, compromising the integrity of the cartilage. The breakdown of extracellular matrix surrounding the chondrocytes allows cytokines and growth factors to readily diffuse through the damaged matrix and into the cartilage more easily, resulting in breakdown and poor repair Krustev *et al.*, 2015). In later stages of OA, a substantial breakdown of collagen occurs, which is thought to be the point of irretrievable cartilage deprivation Mbvundula *et al.*, 2005). Due to the low regeneration capacity of cartilage and the lack of disease-modifying therapy, there is a strong need to develop new therapeutic strategies that could improve chondrocyte function in OA.

There is evidence that cannabinoids have chondroprotective activity and this may aid the development of novel treatments for OA Dunn *et al.*, 2012). Cannabinoids have been demonstrated to have anti-inflammatory effects and to have a positive influence, protecting against cartilage degradation in inflamed arthritic joints Mbvundula *et al.*, 2005*;* McDougall *et al.*, 2008).

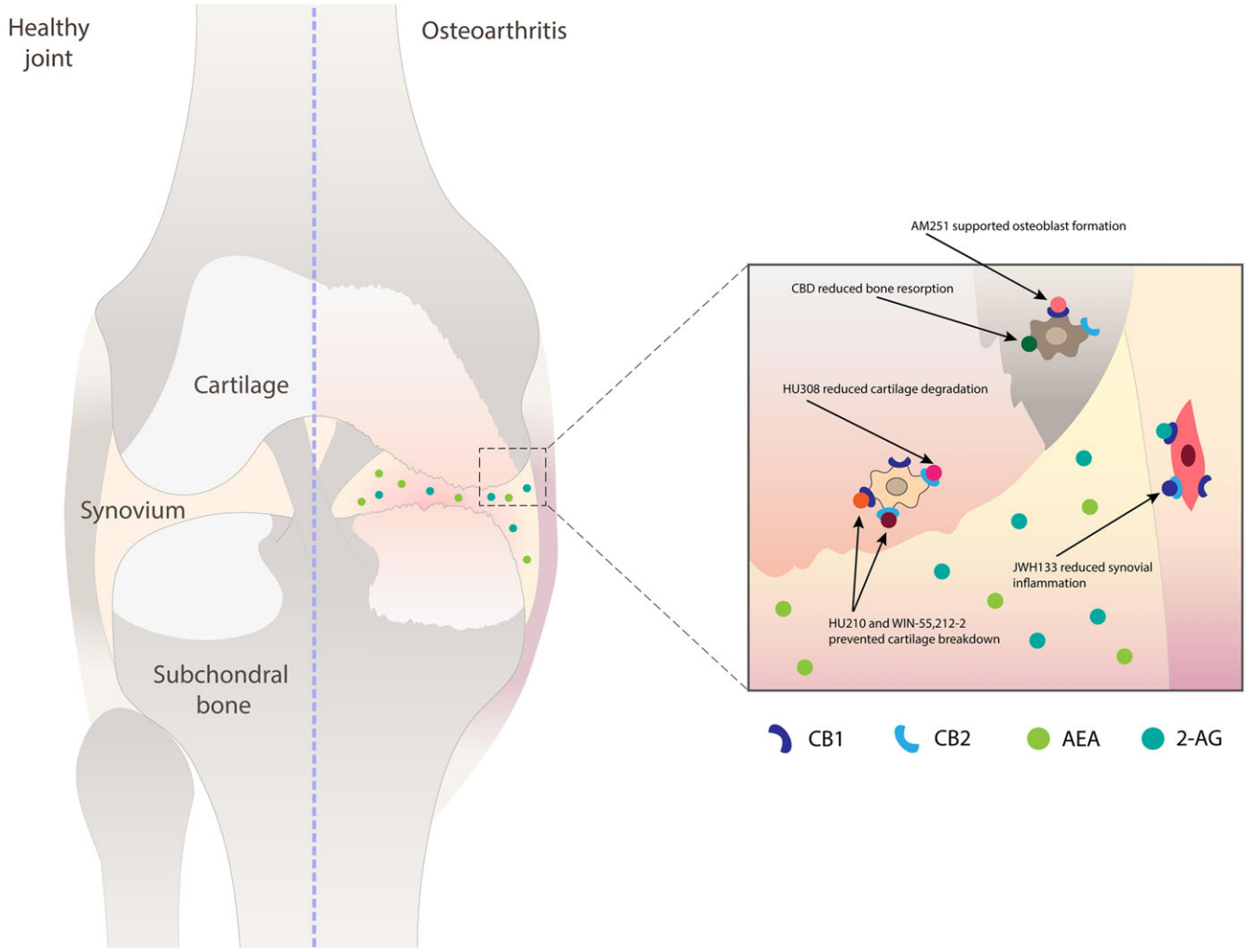
Chondrocytes extracted from OA-affected joints were shown to express both CB_1 and CB_2 receptors even in degenerated tissues, demonstrating that these cells could respond to cannabinoids. Furthermore, studies have shown that both endocannabinoids and synthetic compounds have a direct chondroprotective effect, resulting in an inhibitory effect on proteoglycan breakdown and cartilage protection Mbvundula *et al.*, 2005). Thus, cannabinoids could be protective against joint degeneration in the development of OA. Cannabinoids that are designed to bind to receptors inhibiting the catabolic and pain pathways within the arthritic joint would bypass psychoactive ef cacy, thus providing a novel insight into the therapy of OA Dunn *et al.*, 2014) Figure 1). The CB_2 receptor has been shown to regulate the susceptibility to OA in mice. Studies using the destabilized medial meniscus model of OA revealed that mice lacking CB_2 receptors demonstrated more severe OA phenotype than wild-type OA mice, and showed an increased predisposition to develop age-related OA. Moreover, a selective CB_2 agonist, **HU-308**, reduced the severity of cartilage degradation in wild-type OA mice but not in CB_2-de cient mice. In addition, pharmacological treatment with HU-308 reduced the severity of OA changes within the whole joint. Additionally, in this study, it was found that cultured *Cnr2 /* chondrocytes produce less proteoglycans *in vitro* than wild type chondrocytes Sophocleous *et al.*, 2015). Additionally, it was shown that **HU-210** and **WIN55212-2** synthetic cannabinoids) directly prevent cartilage degradation *via* a CB_1- and CB_2-dependent mechanism Mbvundula *et al.*, 2006*;* Dunn *et al.*, 2014). WIN55212–2, which acts on CB_2 receptors, was also found to inhibit the activity of **ADAMTS-4** disintegrin and metalloproteinase with thrombospondin motifs 4) in articular chondrocytes treated with **IL-1** , thus preventing cartilage breakdown in OA Dunn *et al.*, 2014).

Other studies have also noted that other receptors may be responsible for the chondroprotective effect of cannabinoids. GPR55 and GPR18, which are known to respond to cannabinoid treatments, have both been shown to be expressed in normal and OA cartilage Goldring and Marcu, 2009). Although there was no connection between GPR55 expression and cartilage degradation, the GPR18 immunopositivity of chondrocytes decreased with disease progression. This may suggest this receptor is involved in cartilage degeneration. In addition, the expression of TRPV1, which is an endogenous AEA receptor, decreased with changes occurring in chondrocytes in OA progression Gavenis *et al.*, 2009). These novel ndings indicating the possible involvement of GPR18 and TRPV1 in cartilage metabolism could broaden the knowledge on the endocannabinoid system in OA and should be investigated further.

Although previous studies have shown that cannabinoids display chondroprotective properties, the expression of cannabinoid receptors within OA cartilage is poorly associated with the degree of degeneration. Moreover, the role of AEA in chondrocyte cell viability is still unclear. AEA has been found to decrease chondrocyte vitality, although this effect was not CB receptor-dependent. Nevertheless, it was also shown to work in synergy with TNFα to cleave caspase-3 and induce apoptosis, thus facilitating cartilage degeneration Gómez *et al.*, 2014). Therefore, it is important to further identify the role of these receptors within normal and OA cartilage to clarify their role in healthy tissue and their potential as possible targets in the treatment of OA Dunn *et al.*, 2016).

Subchondral bone in the pathogenesis of OA: role of cannabinoids

The widely held belief that OA is a disease of cartilage has been a subject of debate. Several investigations have led to the hypothesis that bone changes may account for joint deterioration and the development of OA. Indeed, individuals with OA exhibit striking increases in bone mass at affected sites, such as the knee and hip, and at non-synovial sites such as the lumbar spine. Individuals with OA also have a high body mass index and exhibit an increase in bone mineral density. The latter may be the result of an abnormally high rate of metabolism of osteoblasts, which is observable *in vivo* and *in vitro*, particularly in subchondral bone Sharma *et al.*, 2013). An increase in bone turnover, the presence of oedema-like lesions in subchondral bone marrow and bone attrition are strong indicators of structural deterioration of the knee in OA Raynauld *et al.*, 2006). Indeed, most current research highlights the fact that all of the joint components and particularly subchondral bone undergo functional and structural changes during OA progression Funck-Brentano and Cohen-Solal, 2015). Given the intimate contact between the cartilage and bone, alterations of either tissue will modulate the properties and function of the other joint component. It is still a matter for debate, however, whether the subchondral bone changes occur at the same time as changes in articular cartilage, and thus are causative, or if, as



Figure

Effects of CB_1 and CB_2 receptor ligands on joint tissue metabolism in OA. During OA, functional units of joints comprising cartilage, subchondral bone and synovium undergo uncontrolled catabolic and anabolic remodelling processes to adapt to local biochemical and biological signals. Modulation of the cannabinoid system can affect the major facets of joint biology. Cannabinoid-based drugs aim at i) reducing cartilage degradation and facilitating cartilage repair; ii) causing osteoclast apoptosis and inhibit osteoclast formation *n v tro*, thus acting as anti-resorption agent; and iii) acting on syniviocytes as immunomodulatory and anti-in ammatory agents. All those effects may be important for the treatment of OA progression. CB_1 and CB_2 receptor activation in OA joints could improve multiple aspects of the disease, including suppression of cartilage degradation, bone remodelling and synovitis, all of which leads to alleviation of joint destruction.

a consequence of cartilage degradation, bone is intimately involved in the initiation and progression of OA, where trauma to the subchondral bone may result in cartilage degeneration Funck-Brentano and Cohen-Solal, 2015). Increased trabecular bone volume with trabecular sclerosis and increased bone turnover are features of OA pathogenesis. Therefore, therapies that target bone may also be effective in OA.

There is compelling evidence suggesting the endocannabinoid system actively participates in bone cell differentiation, survival and function. The identi cation of both CB_1 and CB_2 receptors in bone mass Idris and Ralston, 2012) suggests that pharmacological modulation of these receptors could be a valuable therapeutic approach in human bone diseases including OA Ofek *et al*., 2006*;* Whyte *et al*., 2012). Endocannabinoids and their receptors have been reported in the skeleton, both in the bone forming cells osteoblasts) and the bone resorbing cells osteoclasts). The CB_1 receptor is highly expressed in skeletal sympathetic nerve terminals Tam *et al*., 2006). In osteoclasts, CB_1 receptor are expressed at low levels, whereas CB_2 receptors are present in high abundance Ofek *et al*., 2006). *In vivo*, CB_2 receptor protein is present in trabecular osteoblasts Lian *et al*., 2004) and their descendants, the osteocytes, as well as in osteoclasts Ofek *et al*., 2006). Osteoblasts and osteoclasts produce the endocannabinoids AEA and 2-AG and express CB_2 receptors Idris and Ralston, 2012). Notably, studies have demonstrated abnormal bone phenotypes in mice lacking either CB_1 or CB_2 receptors, and these phenotypes varied with age, gender or genetic background Idris *et al*., 2005). The endocannabinoid

system has been implicated in maintaining bone homeostasis in a number of ways; for example, CB_2 knockout mice showed accelerated age-related osteoporosis, whereas activation of CB_2 receptors increased bone formation markers Valverde *et al*., 2005). The endocannabinoid system appears to regulate bone mass by signalling *via* peripheral CB_2 receptors in both osteoblasts and osteoclasts.

There is increasing evidence to support the existence of additional cannabinoid receptors non-CB_1 and non-CB_2) both in the CNS and the periphery, further veri ed in CB_1 and CB_2 receptor knockout mice Valverde *et al*., 2005). Indeed, some actions of certain cannabinoid ligands are mediated by other receptors, TRPV1, GPR55 and GPR18. Recently, it has been reported that CB_1, CB_2 receptors and other receptors related to the cannabinoid system, TRPV1 and GPR55, are also involved in bone metabolism Ofek *et al*., 2006*;* Whyte *et al*., 2012). Among the factors of importance for bone remodelling process, the molecular triad NF B ligand RANKL)/RANK/osteoprotegerin has emerged as an essential player not only in bone formation but also in bone resorption processes; for example, osteoblast cultures from CB_1 knockout mice expressed less RANKL cytokines for osteoclast formation), thus reducing the potential for bone resorption Idris *et al*., 2009). Cell- and tissue-based studies indicate that endocannabinoids influence bone remodelling by acting on CB_1 and CB_2 receptors expressed on bone cells.

However, because cannabinoids interact with other receptors, their potential role in the mechanism of bone remodelling needs more widespread research. Several studies have shown the presence of the classical cannabinoid receptors CB_1 and CB_2 and of GPR55 in bone cells, suggesting their possible role in bone remodelling processes during OA Idris and Ralston, 2012). Indeed, the pharmacological and genetic manipulation of CB_1, CB_2 and GPR55 receptors can suppress bone resorption, increase bone mass and prevent bone loss. The synthetic cannabinoid **AM251**, a CB_1 receptor antagonist/inverse agonist, and the genetic deletion of CB_1 receptors have both been shown to decrease osteoblast differentiation from bone marrow-derived cells Idris *et al*., 2009). Several studies highlight the therapeutic value of CB_2 receptor agonists in bone tissue formation and mineralization Idris *et al*., 2008). The non-psychoactive cannabinoid, **cannabidiol** CBD), can be an effective oral anti-arthritic therapy Malfait *et al*., 2000). Moreover, CBD can reduce bone resorption, possibly by modulating GPR55 receptor signalling Dunn *et al*., 2016). However, GPR55 s actions are still unclear, and further research is essential to foster a deeper understanding of its role in bone metabolism, especially under pathophysiological conditions. Our recent studies demonstrate the contribution of the CB_2 receptor to the cytoprotective effects of AEA on primary microglia cells in LPS-induced neuroinflammation Malek *et al*., 2015*b*). We have also shown the involvement of GPR18 in these effects and suggest possible crosstalk between CB_2 and GPR18 receptors Malek *et al*., 2015*b*). Bone remodelling has been associated with modi cation of sensory afferent innervation in bone cancer patients. Hence, similar mechanisms may also play a role in OA, where a possible link between the CNS and bone formation may be present. Thus, GPR18 may also be a new attractive target in bone metabolism regulation.

Last but not least, the co-expression of TRPV1, CB_1/CB_2 receptors and AEA metabolic enzymes was found in both human osteoclast cultures and in native osteoclasts from human bone biopsies Rossi *et al*., 2009). Moreover, agonist-evoked calcium entry indicated that the TRPV1 receptor is functionally active *in vitro*. Consistently, biomolecular and functional experiments showed that **resiniferatoxin**, a selective TRPV1 receptor agonist, increased the expression and the activity of two speci c osteoclast biomarkers, TRAP and cathepsin K. The evidence that cannabinoid and vanilloid receptors are co-expressed in human osteoclasts suggests that they might crosstalk to modulate the intrinsic balance of bone mineralization and resorption by different actions of AEA through TRPV1 and cannabinoid receptors.

The most important feature of OA is that structural changes in the joint involve the major constituents of articulation, including subchondral bone. Whether these changes affect cartilage before reaching subchondral bone or *vice versa* is still a matter of debate. A resolution of this question is likely to modify what is known about this disease and any potential treatment.

Cannabinoids in synovial tissue

OA has long been considered a 'wear and tear disease, implicating degeneration connected with the ageing process as its primary and sole cause. Currently, a literature review suggests that inflammation may play a pivotal role in the aetiology of OA. The discovery that many soluble mediators of inflammation, such as cytokines, actively participate in shaping the clinical situation of OA patients led to the rst steps of an 'inflammatory theory of the disease Goldring, 2000a). Moreover, recent experimental data have shown that inflammatory processes may underlie the initiation and prolongation of OA, thereby opening a way to consider inflammation as a crucial factor in the disease. The classi cation of OA as a non-inflammatory arthritis is an unfortunate consequence of early observations noting fewer leukocytes in OA synovial fluid compared with that of rheumatoid arthritis RA), which is indisputably an inflammatory condition. Similarly, early studies in RA, using OA tissues and fluids as a reference, reported dramatically increased levels of inflammatory proteins in RA Smith *et al*., 1997), which reinforced the notion that OA is not associated with inflammation. Moreover, in these initial studies, a similar histopathology between the so-called post-traumatic synovitis and OA has been described, again suggesting the importance of mechanical stress in progressive loss of cartilage Soren *et al*., 1976). Currently, OA is believed to be a very complex multifactorial disease that involves low-grade inflammation in cartilage and synovium and results in the loss of joint structure and progressive deterioration of cartilage. In April 2015, OA Research Society International con rmed a new understanding of the disease by publishing an updated de nition of the disease as follows: OA is a disorder involving moveable joints characterized by cell stress and extracellular matrix degradation initiated by micro- and macro-injury that activates maladaptive repair responses including pro-inflammatory pathways of innate immunity. The disease manifests rst as a molecular derangement abnormal joint

tissue metabolism) followed by anatomical, and/or physiological derangements characterized by cartilage degradation, bone remodelling, osteophyte formation, joint inflammation and loss of normal joint function), that can culminate in illness Kraus *et al*., 2015). It is additionally becoming clear that many of the physiological ailments of ageing are at least associated with, if not directly related to, chronic low-grade inflammation. Given its role in numerous chronic diseases, chronic inflammation should now be considered a key driver of progressive degeneration in OA joints. Increasingly, attention has turned to the role of the synovium in OA, as it is now proposed that synovitis is common and associated with OA pain.

A set of data from the Chapman group identi ed that the cannabinoid receptor system present in the synovium may be an important therapeutic target for the treatment of pain and inflammation associated with OA Richardson *et al*., 2008). These authors not only demonstrated the presence of an active endocannabinoid system, including endocannabinoids, entourage compounds, CB_1 and CB_2 receptors and fatty acid amide hydrolase **FAAH**), in the knee synovia of patients with end-stage OA and RA but also provided strong support for functionally coupled cannabinoid receptors in the broblast-like cells derived from synovia from OA and RA patients. In a series of experiments, the non-selective cannabinoid receptor agonist HU-210 produced phosphorylation of the downstream signalling kinases, indicating an increase in ERK and p38-MAPK activity. These pharmacological studies provide strong support for functionally coupled cannabinoid receptors in the broblast-like cells derived from synovia of OA patients Richardson *et al*., 2008).

The synovium is the most important site of cytokine production in arthritis, and synovial cells from arthritic animals are known to spontaneously produce large amounts of TNFα when cultured *in vitro* Malfait *et al*., 1998). This broad family of secreted small proteins is crucial in cell communication during inflammation. A board spectrum of pro-inflammatory cytokines in the synovial fluid of OA patients can be detected easily Schlaak *et al*., 1996). *In vitro* studies have demonstrated that exposure of naive synoviocytes to pro-inflammatory cytokines leads to an up-regulation of CB_1 receptors but a down-regulation of CB_2 receptors McPartland, 2008). Additional *in vitro* studies showed that synovial cells from mice with inflammatory arthritis produced large amounts of TNFα Malfait *et al*., 2000), which is the main contributor to inflammation in arthritis.

A signi cant amount of research has clari ed the contribution made by the synovium to RA, but more research is needed to elucidate its contribution in OA pathology, which has long been underestimated. In fact, the synovium as a whole has received less attention than have the other joint tissues, partly due to the former s relative inaccessibility and fragility in a typical rodent model. A well-established contributor of joint inflammation is the in ltration of synovial macrophages. Macrophages express CB_2 receptors, and cannabinoid receptors are expressed on neuronal cells. Therefore, there is a scope for a complex pattern of cannabinoid interactions within the synovium and surrounding joint tissue.

The enthusiasm for using cannabinoids as medicines seems to go through phases. The separation of CB_1 and CB_2 receptors implies that the activation of CB_2 receptors will lack psychotropic effects, so considerable efforts have gone into selective ligands for this receptor in particular. Fukuda *et al*. 2014) tested the ef cacy of the selective CB_2 agonist **JWH133** against broblast-like synoviocytes FLS) inflammation. The authors encouraging results show that *in vitro* the FLS culture from RA patients produces **IL-6**, **MMP3** and **chemokine C-C motif) ligand 2 CCL2)** in response to TNFα stimulation. This is interesting at a number of levels. IL-6 is known to induce pain Alvarez and Levine, 2014) along with MMP3 having a role in matrix turn over, which may diffuse from the synovium into cartilage in parallel to MMP13 Moore *et al*., 2000). Fukoda *et al*. also showed that JWH133 was able to inhibit CCL2 expression, which is a chemokine involved in the recruitment of monocytes, macrophages, T-cells and dendritic cells to sites of inflammation. Inhibitory effects of CB_2 receptors on synovial broblasts in RA were also reported by Gui *et al*. 2014). In FLS isolated from RA patients, pro-inflammatory mediators up-regulate the expression of CB_2 receptors, negatively regulating the production of pro-inflammatory cytokines and MMPs. These data suggest that the CB_2 receptor may be a potential therapeutic target of RA. Thus activation of the cannabinoid receptor system might be an adaptation to the pro-inflammatory environment in RA and might help resolve inflammation, although a question arises of whether this is true in OA.

Recently, anti-inflammatory effects of AEA in synovial cells both from RA and OA-patients were reported to be mediated by TRPV1 in a **COX-2**-dependent manner. A possible anti-inflammatory mechanism involves the down-regulation of MAPK signalling by AEA. Activation of the endocannabinoid system can be bene cial in arthritis, and manipulating this system especially by using a combination of COX-2 and a FAAH inhibitor) might be a promising strategy to reduce erosions and inflammation in arthritis Lowin *et al*., 2015). This emphasizes the importance of elucidating the involvement of the endocannabinoid signalling in OA in further detail. Future research should also concentrate on phytocannabinnoids. CBD has very strong anti-inflammatory properties. Synovial cells from mice treated with exogenous CBD produced signi cantly less TNFα in culture Malfait *et al*., 2000). CBD had a dose-dependent therapeutic effect on disease progression in mice with inflammatory arthritis. The CBD treatment not only suppressed clinical signs of the disease, but no obvious side effects were noted with chronic treatment Malfait *et al*., 2000). Furthermore, CBD treatment protected the hind paws from these mice from joint destruction in both acute and chronic disease states when compared to control Malfait *et al*., 2000).

The above evidence supports the possibility of using cannabinoids as a novel therapeutic target and/or potential drug in the progression of rheumatic disease states. The question arises as to how to modulate the EC system for the treatment of OA. We believe that in light of data investigating the role of the endocannabinoid system in RA, its modulation will be a treatment for OA as well. CB_1 antagonists might reverse the metabolic alterations associated with OA, while activation of CB_2 receptors might be bene cial in patients by down-regulating cytokine production. As cytokines are highly important in OA, they are promising candidate

biomarkers for disease diagnosis and prognosis and are subjects for modulation by endocannabinoids. There is a great need for further work on this issue, which, to a certain degree, may be achieved by undertaking future research.

Summary

The originating event accountable for the development of OA is still not known. It is highly probable that, disregarding the initial events, all three tissues mentioned here contribute to OA progression. It was shown that the factors released by osteoblasts might modify chondrocyte differentiation and proliferation Sanchez *et al*., 2005). Although the role of the cannabinoid system in joint functioning is well documented in preclinical studies, the evidence from clinical studies is still insuf cient to conclude what effects cannabinoids have on OA progression and treatment. OA, therefore, may not be a single disease but may be the outcome of different pathological processes. In one case, OA may be caused by a process restricted to the cartilage, while in another, synovial involvement may be more important with apparent inflammation. Understanding these possible cellular and molecular interactions, particularly between cartilage and bone leading to the progression of OA, represents an important issue in directing treatment to either compartment bone or cartilage) and may provide a circuit breaker in OA to prevent or slow the progression of this condition**.** The expression of cannabinoid receptors was higher in chondrocytes than it was in osteocytes, suggesting that chondrocytes may be more responsive to cannabinoid treatment, thus facilitating the chondroprotective effects. Despite this, none of the clinical trials conducted so far have included observations on the effect of cannabinoids on the progression of cartilage, bone or synovium impairments in OA. Likewise, limited evidence is available to support the medical use of cannabinoids in OA-related pain. The most notable clinical studies aimed at modulating the endocannabinoid system for the alleviation of OA pain were conducted with the irreversible FAAH inhibitor, **PF-04457845** Huggins *et al*., 2012). This compound showed excellent tolerability and elevated the plasma levels of AEA. However, despite the analgesic properties of FAAH inhibition in rodent models of OA pain Schuelert *et al*., 2011), it failed to provide analgesia in knee OA patients Huggins *et al*., 2012). There are possible explanations for the lack of an analgesic effect, but further studies of this type are necessary for details, see Malek and Starowicz, 2016). The main problem in the translation of results from animal models to clinical applications might be the mechanisms underlying OA. As the mechanisms are still not fully understood, they limit the direct conversion of the data from preclinical work and raise a question of how OA patients can be divided into diverse subgroups based on the origin and development of the disease.

To sum up, as *in vitro* studies reveal promising data about a potential role of the endocannabinoid system in the modulation of the mechanisms underlying structural pathology during OA, including their possible involvement in the repair mechanisms, further studies exploring the overall *in vivo* effects of these agents in preclinical models are necessary to support the potential interest of cannabinoids for halting the progression of this disease.

Nomenclature of Targets and Ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY Harding *et al*., 2018), and are permanently archived in the Concise Guide to PHARMACOLOGY 2017/18 Alexander *et al*., 2017a,b,c).

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Con ict of interest

The authors declare that there are no conflicts of interest.

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