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Review

Osteoarthritis year in review 2020: biology

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SUMMARY

This year in review about osteoarthritis biology highlights a selection of articles published between the 2019 and 2020 Osteoarthritis Research Society International (OARSI) World Congress meetings, within the field of osteoarthritis biology. Highlights were selected from PubMed searches covering osteoarthritis (OA) cartilage, subchondral bone, synovium and aging. Subsequently, a personal selection was based on new and emerging themes together with common research topics that were studied by multiple groups. Themes discussed include novel insights into the inflammatory changes during OA, with a number of noteworthy publications concerning the role of macrophages in healthy and osteoarthritic joints. Next, the application of mesenchymal stem cells as OA-dampening therapy is discussed, including possible ways to improve their efficacy by pre-treatment. Other significant themes including treatment of OA with metformin, enhancing autophagy to alleviate OA and the involvement of the gastro-intestinal microbiome in development of OA symptoms and structural damage are discussed. An effort was made to connect the seemingly distant topics from which the overarching conclusion can be drawn that over the last year promising breakthroughs have been achieved in further understanding the biology of OA development and that new therapeutic possibilities have been explored.

Search criteria and selection process

The basis of this year in review of osteoarthritis (OA) biology was formed by a personal selection of articles investigating biological mechanisms of OA that were identified using PubMed searches. Selected articles were published between the 2019 and 2020 Osteoarthritis Research Society International (OARSI) World Congress Meetings. PubMed searches were done using combinations of “osteoarthritis” and “cartilage”, “synovium”, “subchondral bone” or “aging”, yielding a total of 1,567 publications. This was reduced to 499 publications after removal of publication dealing with imaging, clinical trials, rehabilitation & outcomes, biomechanics, biomarkers and genetics, genomics and epigenetics, which are dealt with elsewhere in this Year in Review series.

This review does not elaborate on all the laudable work that has been published over the last year, but rather aims to highlight common research topics that were studied by multiple groups and significant breakthroughs in the field of osteoarthritis research. An overview of the presented manuscripts can be consulted in Fig. 1.

Novel insights into inflammatory processes

Relatively low-grade inflammation is present in the majority of OA patients, characterized by synovitis and a pro-inflammatory/catabolic state of the chondrocytes. Grandi *et al.* profiled healthy and OA articular cartilage using single cell mass cytometry and identified 20 chondrocyte clusters of which 13 showed a different abundance between healthy and OA patients[1]. Inflammation-amplifying and inflammation-dampening clusters were present, both relatively rare and only comprising 0.5–1.5% of all chondrocytes. The inflammation-amplifying population was consistently increased in OA patients and expressed IL1R1 and TNFRII and showed exclusive signaling via JNK and SMAD1/5 pathways compared to other clusters. The inflammation-dampening population was positive for CD24 and positively correlated with the inflammation-amplifying population abundance. Interestingly, a combined JNK-inhibiting and CD24-increasing treatment showed a strong decrease in the chondrocyte production of various chemokines, indicating the potential of modulating the balance between these populations to dampen joint inflammation.

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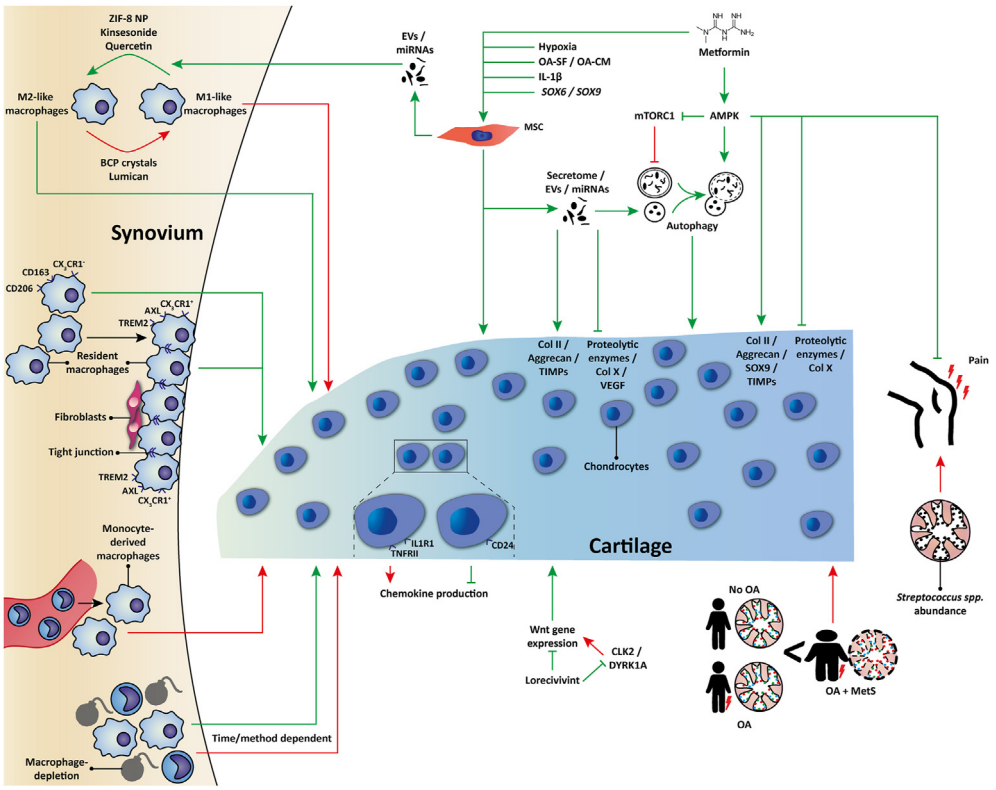

Fig. 1

Schematic summary of the findings of the osteoarthritis biology studies featured within this 2019–2020 review. This is a simplified scheme of the publications as discussed in this review, showing connections between the reported findings. Green indicators represent a supportive effect (protective against OA), while red indicators represent a detrimental effects (promoting OA). Pointed arrows indicate stimulatory effects, whereas blocked lines indicate inhibitory effects. AMPK: 5′ AMP-activated protein kinase, AXL: AXL receptor tyrosine kinase, CD163: cluster of differentiation 163, CD206: cluster of differentiation 206, CLK2: CDC like kinase 2, Coll II: type two collagen, Coll X: type 10 collagen, CX3CR1: C-X3-C motif chemokine receptor 1, DYRK1A: dual specificity tyrosine phosphorylation regulated kinase 1 A, EV: extracellular vesicle, IL-1β: interleukin one beta, MetS: metabolic syndrome, miRNA: micro ribonucleic acid, mTORC1: mammalian target of rapamycin complex 1, OA: osteoarthritis, CM: conditioned medium, SF: synovial fluid, SOX6: SRY-box transcription factor 6, SOX9: SRY-box transcription factor 9, TIMP: tissue inhibitor of metalloproteinase, TREM2: triggering receptor expressed on myeloid cells 2, VEGF: vascular endothelial growth factor, ZIF-8 NPs: zeolitic imidazolate framework-8 nanoparticles.

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Deshmuck *et al.* showed, that the novel small-molecule lorecivivint exhibits anti-inflammatory capacities by inhibiting NFκB and STAT3 signaling, which was associated with increased chondrocyte differentiation and decreased cartilage degradation. They showed that lorecivivint inhibited CLK2 and DYRK1A, therewith modulating the Wnt signaling pathway downstream and independent of ß-catenin at a posttranscriptional level. Together, although further clinical trials are necessary to show its efficacy in meeting the endpoint criteria, this compound may present as a promising first DMOAD[2]. However, lorecivivint failed to reach the primary end point, a significant improvement in the Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC) pain score after 13 weeks of treatment as compared to placebo, in a recent phase IIa trial[3].

The low-grade synovitis is mainly characterized by activation of innate immune effector cells, in which macrophages emerged as key dysregulatory cell type. Although it has been previously shown that T cells are present in the synovium, OA is generally not considered an autoimmune disease[4].

Still, Camacho-Encina and co-workers described the discovery of a signature of six autoantibodies that associated with future disease incidence[5]. Autoantibodies against methionine adenosyltransferase two subunit beta were further validated as discriminating marker. The authors warrant future studies, based on the biological involvement of this protein in various processes that are involved in OA development.

Macrophages are generally considered critical mediators for the maintenance of tissue homeostasis and, consequently, are thought to be involved in the pathology and symptomology of OA once dysregulated. Over the last year, several excellent studies have been published that add to our body of knowledge about these cells and their behavior in the joint.

A comprehensive study by Culemann *et al.* showed that synovial macrophages consist of multiple subgroups using a broad fate

mapping approach[6]. This revealed that joint resident synovial macrophages can be subdivided into CX_3CR1^+ cells, populating the uppermost synovial layer on top of the fibroblast-like synoviocytes, and CX_3CR1^- interstitial macrophages. CX_3CR1^+ macrophages form a dense physical barrier that secludes the joint cavity from the surrounding synovium and as such controls the onset of inflammation to protect intra-articular structures. These CX_3CR1^+ cells are thought to derive from early embryonic hematopoiesis and to maintain its numbers independent of blood monocytes. Interestingly, proliferating CX_3CR1^- cells in the deeper synovial layers were shown to repopulate the CX_3CR1^+ lining cells with a half-life of approximately 5 weeks. Deeper phenotyping showed that $MHC^+CX_3CR1^-$ cells proliferated and further differentiated into RELM-$\alpha^+CX_3CR1^-$ interstitial macrophages that additionally expressed *Mrc1* and *Cd163*, associated with M2-like macrophage, and CX_3CR1^+ lining macrophages. The authors furthermore show that the CX_3CR1^+ lining macrophages formed tight-junctions and expressed immunomodulatory markers, such as *Trem2* and *Axl*, indicative of an anti-inflammatory M2-like phenotype, with limited response to inflammation and largely conserved their naïve state upon pro-inflammatory triggering.

Induction of experimental arthritis led to additional clusters of pro-inflammatory monocyte-derived macrophages and disintegration of the protective barrier as the result of immune complexes but not after depletion of PMNs or $Ly6C^{high}$ inflammatory monocytes, showing that this was not the result of myeloid cell influx. Although this study focused on rheumatoid arthritis, it is valuable to the OA field. It shows the difference between the pro-inflammatory monocyte-derived macrophages and the immunomodulatory resident macrophage population forming a protective barrier in the lining, the function of which during OA development could be interrogated in future studies.

This manuscript sheds new light on, but also further complicates, the interpretation of results from studies in which research groups determined whether joint tissues could benefit from systemic or local macrophage-depletion. Earlier studies showed that macrophage depletion before induction of collagenase-induced OA protected against cartilage degradation and osteophyte development[7,8]. In a more recent study by Wu *et al.* conditionally and systemically depleted macrophages after destabilization of the medial meniscus (DMM) induction in mice on a high fat diet[9]. The Macrophage Fas-Induced Apoptosis (MaFIA) mouse was used, from which $Csf1r^+$ cells can be conditionally depleted by administration of AP20187, which means that not only macrophages, but also pro-inflammatory monocytes are depleted[10]. Moreover, given the results from the study by Culemann, only pro-inflammatory infiltrating monocyte-derived macrophages and the *Csf1r*-expressing $MHC^+CX_3CR1^-$ interstitial macrophage are depleted, whereas the protective CX_3CR1^+ and RELM-$\alpha^+CX_3CR1^-$ macrophage populations will not be targeted because these are devoid of *Csf1r* expression. Surprisingly, therefore, Wu *et al.* showed increased synovial inflammation with increased numbers of lymphocytes and neutrophils, whereas OA pathology was not ameliorated. One explanation might be that the protective CX_3CR1^+ lining macrophages could not be replaced because the $MHC^+CX_3CR1^-$ cells that they originate from were depleted.

A follow-up study by Bailey and coworkers described that macrophage depletion by either intra-articular clodronate-laden liposomes, which mainly kill lining macrophages, or the MaFIA mouse did not decrease synovitis after induction of a closed articular fracture of the tibial plateau[11]. Interestingly, macrophage depletion in the MaFIA mice 2 days before induction even increased synovitis. Moreover, significantly more macrophages were present 7 days post-fracture in the synovial stroma of MaFIA mice with macrophages depleted at the day of fracture induction or 2 days thereafter. These macrophages predominantly had an M1-like phenotype. Interestingly, no differences in numbers of lining macrophages could be observed between groups.

Together, these studies show that the technique used for macrophage depletion might affect the outcome. Moreover, the timing of depletion is of crucial importance because of the different functions that tissue-resident and infiltrating monocyte-derived macrophages have in immune regulation over time.

A number of other publications have focused on ways to influence macrophage polarization. Historically, macrophages were dichotomized into pro-inflammatory/catabolic M1 macrophages and M2 macrophages that mainly secrete anti-inflammatory and pro-resolving/anabolic molecules. Although it is now more generally accepted that these represent the extremes of a continuous spectrum, authors often still refer to M1-like and M2-like macrophages while discussing e.g., the direction of treatment effects. A study conducted by Mahon and co-workers showed that basic calcium phosphate crystals cause an M1-like macrophage differentiation, accompanied with a shift towards glycolytic energy metabolism, which has been shown to control inflammatory macrophage functions[12,13]. Another study demonstrated that the cartilage extracellular matrix protein lumican was increased in the synovial fluid (SF) from OA patients and augmented LPS-induced TLR4 signaling, which associated with increased expression of M1-like and decreased expression of M2-like macrophage markers[14].

In contrast, Zhou *et al.* described that zeolitic imidazolate framework-8 (ZIF-8) nanoparticles (NPs), loaded with S-methylisothiourea hemisulfate salt to inhibit nitric oxide synthase and catalase to catalyze O_2 from H_2O_2, steered macrophage polarization towards the M2-like phenotype and inhibited cartilage degeneration after induction of anterior cruciate ligament transection (ACLT) in mice[15]. Two further studies showed that the plant extracts kinsesonide and quercetin directed macrophage polarization towards the M2-like phenotype[16,17].

Together, these studies lend further support for macrophages as key mediators in OA-associated inflammation and furthermore show that modulating these cells or intervening with factors that modify their phenotypic state is still considered a promising approach to slow down OA development.

OA treatment with mesenchymal stem cells

Although mesenchymal stem cells (MSCs) have been predominantly tested with the aim to repair cartilage, in more recent studies they have been applied because they produce anti-inflammatory mediators and induce reparative properties in a paracrine manner. The large 'Adipose derived stromal cells for osteoarthritis treatment' (acronym: ADIPOA) consortium studied the application of adipose-derived stromal cells (ADSCs) during OA development and multiple resulting papers showed that intra-articular ADSC injection ameliorated pathology after induction of experimental OA in various species[18,19]. ADSCs share similar properties with, but are more easily obtained than bone-marrow derived MSCs. Their exact working mechanisms to reduce OA development remains largely elusive.

Because evidence accumulated that the protective effects of MSCs were not the result of direct chondrogenesis but rather rely on paracrine effects, Chen and co-workers tested whether the secretome from MSCs protected against OA development in a rat OA model induced by ACLT and DMM[20]. Injection of conditioned medium (CM) made from human MSC cultures resulted in decreased cartilage degeneration and reversed changes in the subchondral bone such as decreased bone volume vs total volume and decreased trabecular number. Moreover, MSC-CM reversed decreased expression of the cartilage matrix fragments aggrecan

and type two collagen, whereas it significantly decreased the expression of MMP13, increased the expression of TIMP1 and reversed the decreased expression of the autophagy markers LC-3 and Beclin-1, coinciding with decreased chondrocyte apoptosis. In agreement with this, a study by Niada *et al.* showed that the ADSC-secretome reduced TNFα-induced chondrocyte hypertrophy and the expression of catabolic markers[21].

Ragni and co-workers conducted high-throughput screening for miRNAs in extracellular vesicles (EVs) secreted by hADSCs cultured under inflammatory conditions that mimic the OA environment in combination with multiplex protein detection[22]. They identified a set of secreted proteins and miRNAs that decreased cartilage matrix degradation, were chondroprotective and anti-inflammatory by steering macrophage polarization towards M2-like cells. Although miRNAs associated with degenerative processes were also identified, scoring the genetic weight showed a predominance of EV-carried miRNAs involved in cartilage and synovium protection.

Previous studies showed that already low numbers of stem cells were effective in ameliorating OA pathology and symptoms, and that their anti-inflammatory effects were boosted by a pro-inflammatory OA environment[23–26]. Because it is being hypothesized that ADSCs depend on chemotaxis to the affected tissues/cells to exert their paracrine effects, a notable study by Manferdini *et al.* showed increased migration under hypoxic conditions, and showed that migration was significantly increased after treatment of ADSCs with OA-SF and OA conditioned medium compared with healthy SF, coinciding with changes in various chemokine receptors on the ADSCs[27]. Another study identified IL-1β as activator of ADSCs that is present in the OA environment, and as such was suggested to be utilized to preactivate the ADSCs before injection[28]. In this line, multiple other studies have focused on pre-treating ADSCs in order to improve their *in vivo* functionality.

A study conducted by Ko *et al.* showed that *SOX6* and *SOX9*-transfected hADSCs showed chondrogenic capacities *in vitro* that were similar to BMP7 and TGF-β-containing cultures[29]. Moreover, intra-articular injection of these cells after medial meniscectomy surgery in goats significantly decreased cartilage degeneration and osteophyte formation, as compared to joints injected with saline or non-transfected hADSCs. Based on the relatively quick disappearance from the joints within 2 weeks after injection, this research group also proposed a model in which the ADSCs exert anti-inflammatory paracrine effects and did not directly differentiate towards articular chondrocytes.

Park *et al.* showed that metformin pre-treatment augmented the anti-inflammatory and anti-pathologic effects of ADSCs[30]. Metformin increased the expression of the anti-inflammatory mediators IDO and IL-10, whereas levels of pro-inflammatory factors including IL-1β, IL-6 and HMGB1 were decreased. Co-culture of ADSCs with human OA chondrocytes using a transwell system showed that metformin-treated ADSCs significantly decreased the expression of various MMPs, type X collagen and VEGF, while increasing TIMP1 and TIMP3 in chondrocytes, whereas non-pre-treated ADSCs did not. Metformin pre-treatment increased the retention time in the inflamed joint, which associated with chondroprotective and pain-reducing effects that were not observed after injection of untreated ADSCs. Interestingly, metformin has additionally been investigated as possible OA treatment independent of MSCs.

Metformin as OA treatment option

Over the last year, multiple research groups have published their results regarding the possible protective effects of metformin on OA development. Metformin is a first-line treatment for type two diabetes based on its glucose-lowering effects via suppressing liver gluconeogenesis, enhancing glucose uptake in muscle and fat tissue and decreasing the uptake of glucose in the intestines[31,32]. Next to inhibition of the mitochondrial respiration complex I, many of the effects of metformin run via activation of the nutrient sensor 5′adenosine monophosphate-activated protein kinase (AMPK)[33]. Reduced activation of the AMPK alpha subunit (AMPKα1) was observed in both mouse and human OA chondrocytes. AMPK activity has been shown to counteract cartilage matrix degradation under inflammatory conditions, whereas in the same line AMPK deficiency resulted in accelerated OA development in mice[34–36].

Several cohort studies previously showed that metformin use could decrease OA progression. Lu *et al.* showed that combined use of metformin and COX-2 inhibitors by individuals with OA and type two diabetes decreased the rate of joint replacement as compared to COX-2 inhibitors alone, whereas Wang and co-workers showed decreased cartilage volume loss in patients with radiographic knee OA and obesity that used metformin as compared to non-users[37,38]. How metformin decreased OA development, however, remained elusive. The studies that have been published over the last couple of months all point in the direction that the advantageous mechanisms of metformin treatment during OA indeed run via activation of AMPK.

Li *et al.* the authors showed that metformin treatment, either starting 2 weeks before or 2 weeks after DMM surgery, significantly reduced the development of articular cartilage degeneration, synovial hyperplasia and osteophyte formation[39]. Since metformin is a known inducer of AMPK, they used AMPKα1-deficient mice and showed that the protective effects of metformin administration on the development of OA pathology was lost, indicating that the chondroprotective effect of metformin on OA pathology was mediated via AMPK signaling. Underlining this, they do provide evidence that metformin significantly reversed the upregulation of catabolic factors, such as *Mmp 3*, *Mmp 13*, *Adamts4* and *Adamts5*, by IL-1β or TNF-α in primary chondrocyte cultures, while increasing the expression levels of anabolic factors like *Col2a1*, *Acan*, *Sox 9*. In line with the *in vivo* findings, the protective outcome of metformin treatment was lost in AMPKα1-deficient mice. Furthermore, metformin treatment could significantly reverse the drop in paw withdrawal threshold after induction of DMM, reflecting reduced pain sensitization. In consonance with the development of pathology and effects on gene expression patterns, effects of metformin on pain sensitization were lost in the absence of AMPKα1.

Another study conducted by Feng *et al.* confirmed the protective effect of metformin against OA development after DMM induction[40]. The authors showed lower OARSI scores and fewer MMP13-positive chondrocytes. Also in this study metformin dose-dependently reversed the IL-1β-induced expression of *Mmp 13* and *Col10a1*, while increasing the expression of *Col2a1* and *Sox 9*. Moreover, metformin treatment decreased the percentage of chondrocytes positive for the senescence marker $P16^{INK4a}$ and dose-dependently caused AMPK activation, mTORC1 inhibition and an increased expression of the autophagy marker LC3. Use of the mTORC1 inhibitor rapamycin decreased the IL-1β-induced MMP13 and increased the Col2a1 expression in primary chondrocytes, while these genes were conversely regulated after addition of the autophagy inhibitor 3-MA. AMPK activity can directly stimulate autophagy by stimulating ULK3 activity and indirectly via the inhibition of mTORC1. In line with previous years, various research groups have published work in which they aimed to increase autophagy in order to slow down the progression of OA[41–43].

A third study, by Li and co-workers showed decreased cartilage degradation after intra-gastric or intra-articular administration of metformin as compared to saline administration after DMM induction[44]. Moreover, they showed a reversal of the decreased paw withdrawal threshold and weight bearing asymmetry after

induction of DMM, indicative of a reduced pain sensation. In agreement with the other studies, metformin reversed IL-1β-induced MMP13, while increasing the expression of type II collagen. Inhibition of AMPKα with dorsomorphin, although dorsomorphin does not specifically inhibit AMPKα, negated the metformin effects.

Of note, various epidemiological studies showed that type two diabetes as independent risk factor for OA development[45,46], hypothesized to be mediated via inter alia high glucose-induced accumulation of advanced glycation end products (AGE), increased pro-inflammatory cytokines and catabolic processes in chondrocytes[47,48], although this association could not be replicated in other studies[49]. Animals in above studies all received a normal chow diet. Owing to the glucose-lowering effects of metformin, it would be interesting to further explore its effects on OA in combination with a carbohydrate-rich Western diet.

Microbiome

Western diet also has been shown to affect conditions such as obesity and related metabolic diseases by changing the gastrointestinal microbiome composition[50]. Gastrointestinal bacteria have been shown to produce a wide range of molecules, including enzymes, short-chain fatty acids and metabolites that can induce inflammatory reactions and increase gut permeability. Increased circulating levels of lipopolysaccharide have been shown in obese individuals, which correlated with macrophage infiltration into the joint and with pain scores and radiographic features of OA[51].

In order to further elucidate the interrelationship between the gut microbiome and OA, Boer *et al.* examined stool samples of a subpopulation of the Rotterdam study as a model to resemble the gastro-intestinal microbiome composition[52]. They observed a significant and replicable association between WOMAC pain scores and the inter-individual *ß*-diversity as evaluated at the genus level, but not the intra-individual *a*-diversity that was lost after correction for BMI, whereas K-L sum-scores representing OA severity were not associated with microbiome diversity. They described a significant association between both relative and absolute abundance of *Streptococcus* spp. and WOMAC pain score, even after correction for BMI, smoking, and alcohol consumption. In addition to dietary intake composition, drug use can have profound effects on the gastrointestinal microbiome. After additional correction for proton pump inhibitor and non-steroidal anti-inflammatory drug use, the association slightly weakened but remained significant. Finally, in an attempt to elucidate the mechanism how an increased *S. spp*. abundance is associated with increased joint pain, the authors evidenced that both WOMAC pain scores and *Streptococcus* abundance significantly associated with knee effusion on knee magnetic resonance imaging (MRI) images. Interestingly, significance was lost after adding WOMAC pain scores to the association model between *Streptococcus* and knee effusion, indicating that the association between *Streptococcus* abundance and knee pain runs via inflammatory processes in the joint.

Another study further substantiated the influence of the microbiome on the development of OA. Whereas Boer *et al.* did not find an association between microbiome diversity and OA severity, a study by Huang *et al.* showed that the microbiome composition can directly influence the course of OA development[53]. They propose a two-hit model for OA pathogenesis where the first hit is provided by disadvantageous microbiome and the second hit by joint damage. The authors collected stool from individuals with OA and metabolic syndrome, individuals with only OA and individuals without evidence of OA. Germ-free mice were orally inoculated with the fecal contents and 2 weeks later the meniscal/ligamentous injury model was induced. Mice receiving fecal bacteria from individuals with OA and metabolic syndrome developed more severe cartilage degradation and synovitis as compared to all other groups, which was associated with significantly increased plasma IL-1β, IL-6 and macrophage inflammatory protein 1α. Furthermore, mice that were inoculated with fecal bacteria from individuals with OA and metabolic syndrome showed decreased RNA expression levels of the tight junction protein ZO-1 and occluding in colon tissue, increased bacterial translocation by the gut epithelium and increased plasma lipopolysaccharide levels as compared to all other groups, indicative of an increased gut permeability. Finally the *Fusobacterium* and *Faecalibacterium* genera positively and Ruminococcaceae_*UCG-013* negatively correlated with OARSI histology score, proteoglycan loss and plasma cytokine concentrations. In contrast to the study by Boer *et al.*, Huang and co-workers did not observe an association between OA and the abundance of *Streptococcus* in the stool of human subjects, whereas this association was borderline significant in the inoculated mice. However, OA readout parameters were different between these studies and the sample size of the study by Huang is relatively low. Moreover, the sampling population in the two studies were different, Caucasian in the study of Boer *et al.* and Han Chinese in the study of Huang *et al.* whereas it has been shown that among other ethnicity and dietary composition strongly affects the microbiome composition[54].

Together, these promising studies show a relatively recent and evolving field of microbiome research in relation to OA development, which warrant for more studies in this direction that take factors such as dietary intake and ethnicity into account.

Conclusion

This review aimed to summarize a selection of articles covering common research themes and exciting new findings in the field of OA research, covering work published between the 2019 and 2020 OARSI World Congress meetings and was by no means meant to cover all commendable research that has been published over the last year.

Firstly, recent publications show that macrophages are still considered key regulatory cells in OA-associated inflammation and that targeting these cells or modifying their inflammatory phenotype is still considered a promising option to treat OA. Novel insights show that various subpopulations of immunomodulatory tissue-resident macrophages are present in the joint, together with inflammatory monocyte-derived macrophages after loss of joint homeostasis. These important findings should be considered while studying ways and timing of macrophage targeting in the future.

A promising approach to target inflammation and decrease the pro-inflammatory state of macrophages is by the intra-articular application of MSCs. Challenges regarding their chondrogenic potential in the harsh OA environment has shifted the focus more towards the immunomodulatory capacities of these cells. Significant findings suggest a paracrine working mechanism via their secretome, possibly with an important function for the cargo of EVs. However, under which conditions MSCs lay down the immunomodulatory imprint that is suggested in many publications remains to be elucidated. These insights will guide the further development of pretreatment options to optimize the MSC efficacy and will allow to apply them to people with an unfavorable joint environment.

Other studies discussed here pointed out that the beneficial effect of the biguanide drug metformin is likely mediated by increasing the autophagy-stimulating nutrient sensor AMPK activity. However, because metformin is predominantly used as a glucose-lowering drug its potential might even be increased under diet-induced or disease-induced hyperglycemic conditions.

Finally significant advances were presented on the involvement of the gastro-intestinal microbiome in the development of OA symptoms and structural pathology. Although the body of evidence still is quite limited for its involvement in OA development, this has been clearly associated with other inflammatory, mainly autoimmune, diseases. Together with the studies that show positive effects of metformin on OA structural damage and progression, this shows that dietary intake/composition likely is an important factor that lowers the threshold for OA development. This holds promise for future OA-preventing strategies since the diet is modifiable, whereas the gastrointestinal microbiome is sensitive to therapeutic interventions.

The overarching conclusion of this review is that a plethora of new and promising avenues of interventions have been investigated. Although some research fields are still at a relatively early stage, where further understanding is required to determine the feasibility of therapeutic targeting, other treatment options are much closer, such as the application of immunomodulatory and reparative MSCs or the inhibition of Wnt signaling. Although not touched upon in this review, it is highly likely that a proper patient stratification based on phenotype or endotype is required to proof treatment efficacy of individual treatment options.

Author contributions

Dr. van den Bosch performed the literature search, selected the themes, wrote and edited the work.

Conflict of interest

No direct or indirect financial or other conflicts of interest are related to this work.

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